Neurology

OFFICIAL JOURNAL OF
THE AMERICAN ACADEMY OF NEUROLOGY

JANUARY--1959
Volume 9 - Number 1

For the epileptic, freedom from seizures means many things: he can work . . . travel . . . and most of all, perhaps, take his place with friends and family without fear. This freedom is possible today because of the ever-increasing knowledge about epilepsy and the improved techniques for its treatment. A major role in this advancement is played by modern antiepileptic drugs. Presented here are five distinguished anticonvulsants that can help you give the epileptic the most precious of all gifts: a normal life. Abbott

the meaning of freedom in epilepsy

ANTICONVULSANTS BY ABBOTT

PEGANONE® (Ethotoin, Abbott) — *A hydantoin of exceptionally low toxicity for* grand mal *and psychomotor seizures.*

PHENURONE® (Phenacemide, Abbott) — *Often effective where other therapy fails in* grand mal, petit mal, *psychomotor and mixed seizures.*

GEMONIL® (Metharbital, Abbott) — *Relatively non-toxic, for* grand mal, petit mal, *myoclonic and mixed seizures symptomatic of organic brain damage.*

TRIDIONE® (Trimethadione, Abbott) / **PARADIONE**® (Paramethadione, Abbott) — *Homologous agents for symptomatic control of* petit mal, *myoclonic and akinetic seizures.*

011167

when you prescribe CARBRITAL®

pentobarbital sodium and carbromal •
in Kapseals® and Elixir form

you prescribe sleep

PARKE, DAVIS & COMPANY • DETROIT 32, MICHIGAN

70459

now, for the first time, **liquid** meprobamate

Equanil®

Suspension

Meprobamate, Wyeth

Wyeth
Philadelphia 1, Pa.

acceptably flavored . . .

your answer to tablet problems in anxiety and tension states

- in children
- in the aged
- in all patients who reject tablet medication

SUPPLIED: Suspension, 200 mg. per 5-cc. teaspoonful, bottles of 4 fluidounces. Also available: Tablets, 400 mg., scored, bottles of 50; 200 mg., scored, vials of 50. WYSEALS® EQUANIL, tablets, 400 mg., vials of 50.

Conforms to Code for Advertising

RELIEVES TENSION—MENTAL AND MUSCULAR

Volume 9 **Neurology** *Number 1*

OFFICIAL JOURNAL OF THE AMERICAN ACADEMY OF NEUROLOGY

OFFICERS

FRANCIS M. FORSTER, M.D., *President*

AUGUSTUS S. ROSE, M.D., *President-Elect*

A. L. SAHS, M.D., *Vice-President*

JOSEPH M. FOLEY, M.D., *Secretary*

CHARLES VAN BUSKIRK, M.D., *Treasurer*

BOARD OF EDITORS

Editor-in-Chief: RUSSELL N. DEJONG, M.D., *Ann Arbor, Michigan*

Associate Editor: MABEL G. MASTEN, M.D., *Miami, Florida*

Editorial Board: PEARCE BAILEY, M.D., *Washington, D. C.*

JOE R. BROWN, M.D., *Rochester, Minnesota*

ROLAND P. MACKAY, M.D., *Chicago, Illinois*

H. HOUSTON MERRITT, M.D., *New York, New York*

JAMES L. O'LEARY, M.D., *St. Louis, Missouri*

E. P. RICHARDSON, JR., M.D., *Boston, Massachusetts*

RICHARD B. RICHTER, M.D., *Chicago, Illinois*

J. PRESTON ROBB, M.D., *Montreal, Canada*

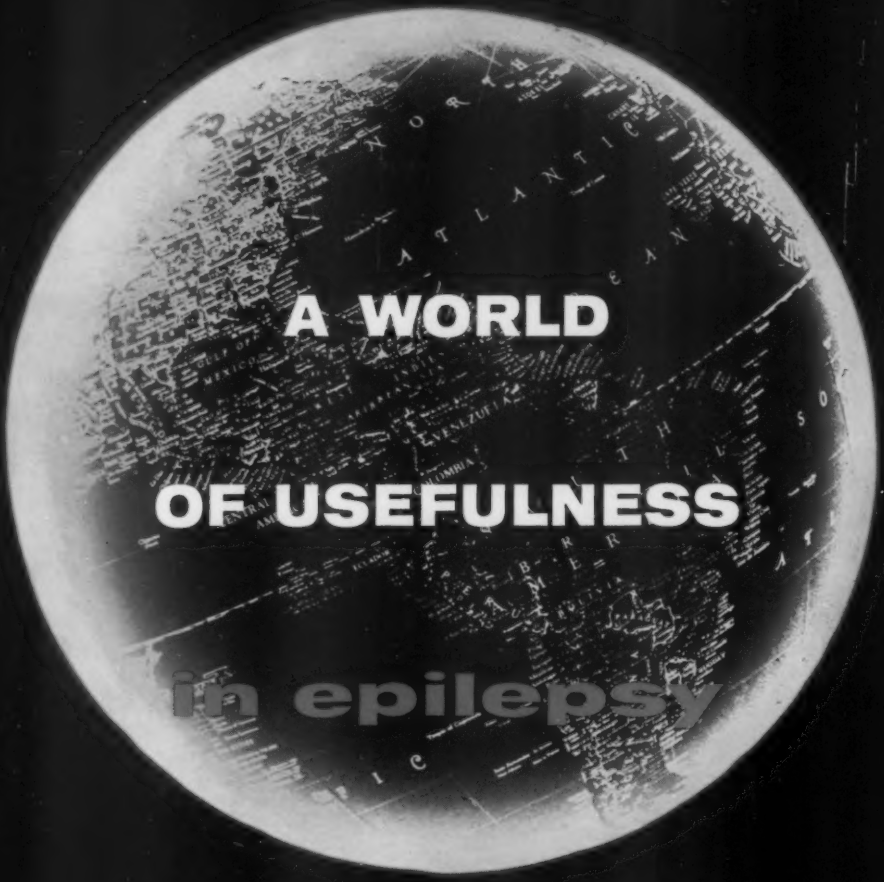
DONALD B. TOWER, M.D., *Bethesda, Maryland*

ALPHONSE R. VONDERAHE, M.D., *Cincinnati, Ohio*

A. EARL WALKER, M.D., *Baltimore, Maryland*

PAUL I. YAKOVLEV, M.D., *Boston, Massachusetts*

NEUROLOGY is published monthly at 84 South Tenth Street, Minneapolis 3, Minnesota, by LANCET PUBLICATIONS, INCORPORATED. Subscription rate: $12.00 a year, $1.00 a copy; outside the United States $13.00 a year, $1.10 a copy. Entered as second class matter at the postoffice at Minneapolis, January 23, 1951.



The world-wide acceptance of "Mysoline" is amply supported by the work of more than 100 investigators in 15 different countries. This effective anticonvulsant for control of grand mal and psychomotor attacks is well tolerated. Side effects, when they occur, are usually mild and transient; and no irreversible toxic effects have been reported. Four years of successful clinical use in the United States further confirms the effectiveness and safety of "Mysoline."

Supplied: 0.25 Gm. scored tablets, bottles of 100 and 1,000.

"MYSOLINE"®

Brand of Primidone

AYERST LABORATORIES · NEW YORK 16, NEW YORK · MONTREAL, CANADA

"Mysoline" is available in the United States by arrangement with Imperial Chemical Industries, Ltd.

Ayerst®

5801

Volume 9 **Neurology** *Number 1*

OFFICIAL JOURNAL OF THE AMERICAN ACADEMY OF NEUROLOGY

JANUARY 1959 • *Contents*

Dissecting Aneurysms of the Carotid Artery Following Arteriography 1
J. F. ROSS FLEMING, M.D., AND ANDREW M. PARK, M.D.

Permeability of Cerebral Blood Vessels in Healing of Brain Wounds 7
J. LEE AND J. OLSZEWSKI

Role of Gamma-Aminobutyric Acid in Electrocortical Arousal 15
E. EIDELBERG, M.D., S. FELDMAN, M.D., AND H. W. MAGOUN, Ph.D.

Radioactive Phosphate Concentration in the Cerebral Cortex 18
LOUIS BAKAY, M.D.

Electroencephalographic Changes in Acute Brain Stem Vascular Lesions 24
WALTER J. FRIEDLANDER, M.D.

Respiratory Effects of Cerebellar and Brain Stem Stimulation in Monkeys 35
HAROLD D. CROSS, M.D., O. WEEMS HOLLOWELL, M.D., AND
JOSE M. R. DELGADO, M.D.

Number of Axons and Myelin Sheaths in Adult Human Medullary Pyramids 42
WILLIAM DEMYER, M.D.

Steroid Suppression of Meningeal Inflammation Caused by Pantopaque 48
JAMES K. SMITH, M.D., AND LIVIA ROSS, M.D.

(Continued on next page)

NEUROLOGY, copyright 1958 by LANCET PUBLICATIONS, INCORPORATED. Title registered United States Patent Office. Louis M. Cohen, Publisher. Mary E. Worthington, Managing Editor. Maurice Wolff, Business Manager, 84 S. 10th St., Minneapolis 3, Minn. ADVERTISING REPRESENTATIVES: *New York 17:* Burt D. Cohen, Lee Klemmer, Bernard A. Smiler, John Winter, 1 E. 42nd St. Telephone: Murray Hill 2-8717. *Chicago 6:* Jay H. Herz, Hugh Gibson, 20 N. Wacker Drive, Suite 1921. Telephone: Central 6-4619. *San Francisco 6:* Duncan A. Scott & Co., 85 Post St., Fifth Floor. Telephone: Garfield 1-7950. *Los Angeles 57:* Duncan A. Scott & Co., 1901 West Eighth St. Telephone: Dunkirk 8-4151.

Contents— *Continued*

Electrogustometric Examinations in Cerebellopontine Tumors and on Taste Pathways 53
BENT KRARUP, M.D.

Hypersensitivity of In Vitro Myasthenic Muscle to d-Tubocurarine 62
JOHN B. DILLON, M.D., AND PHIROZE B. SABAWALA, M.D.

Akinesia in Parkinson's Disease 65
ROBERT S. SCHWAB, M.D., ALBERT C. ENGLAND, M.D., AND ELIZABETH PETERSON

Use of Human Tissue Culture Cells To Attempt Isolation of Virus from Multiple Sclerosis Patients 73
GEORGE E. SEIDEN, M.D.

INFORMATION FOR *Authors*

NEUROLOGY, the official publication of the American Academy of Neurology, has been instituted to provide a medium for prompt publication of articles dealing with neurology and its cognate fields and to stimulate individual investigation in these various fields.

Manuscripts of papers read at meetings of the Academy must be filed with the secretary at the time of the meeting, and will be published if acceptable to the editorial board. They are the property of the Academy, and authors who wish to publish them elsewhere must first request release.

Manuscripts submitted for publication (other than those read at meetings of the Academy) should be sent to Dr. Russell N. DeJong, University Hospital, Ann Arbor, Michigan. Books for review and business correspondence should be addressed to NEUROLOGY, 84 South Tenth Street, Minneapolis 3, Minnesota. News items related to the American Academy of Neurology or to the field of neurology in general are to be sent to Dr. Pearce Bailey, 4725 Sedgwick, N.W., Washington 16, D.C.

Manuscripts are accepted for publication on condition that they are contributed solely to NEUROLOGY. They must be typewritten, double spaced, and only the original copy should be submitted. Illustrations should be drawn in India ink on white paper with clear lettering. Photographs should be glossy prints. Tables should be typed on separate sheets. The title of the article, name of author, and number of figure should be placed on the back of every illustration, and the top should be designated. Legends for figures should be typed and are not to be attached to the illustrations. If the number of illustrations or tables is excessive, the author will be asked to defray a part of the cost.

References should be designated in the text by superior number, and should be listed at the end of the article in numerical sequence. They should be typed in the following manner:

Journal reference: 1. FRANKLIN, C. R., and BRICKNER, R. M.: Vasospasm associated with multiple sclerosis. Arch. Neurol. & Psychiat. 58:125, 1947.

Textbook reference: 2. WECHSLER, I. S.: A Textbook of Clinical Neurology, ed. 3. Philadelphia, W. B. Saunders Company, 1935, pp. 194-199.

Abbreviations should follow the style of the Quarterly Cumulative Index Medicus.

Galley proofs will be sent to authors for prompt correction. There will be no charge for corrections unless excessive alterations are made. A schedule of charges for reprints will accompany the galley proof.

improved functional control

IN PARKINSONISM

Parsidol provides effective and safe control of tremor and muscular rigidity in Parkinsonism.[1,2] As functional decline is retarded, the increased freedom of movement permits many activities formerly not even attempted.[2]

Parsidol improves the patient's emotional perspective as his physical coordination and dexterity return. Though effective by itself, Parsidol is also compatible with most other anti-parkinsonian drugs. Side effects are minimal. Most patients respond optimally to a maintenance dosage of 50 mg. q.i.d.

1. Doshay, L. J. et al.: J.A.M.A. 160:348 (Feb.) 1956
2. Berris, H.: J.-Lancet 74:245 (July) 1954.

PARSIDOL®

brand of ethopropazine hydrochloride

WARNER CHILCOTT

MORRIS PLAINS, N.J.

NEW STELAZINE* EFFECTIVE IN

IN CHRONIC PSYCHOTIC PATIENTS REFRACTORY TO OTHER THERAPIES

"It is obvious that we have in ['Stelazine'] a very effective drug which often produces results where other treatment has failed."

Gunn, D.R.: The Role of Trifluoperazine in the Treatment of Refractory Mental Patients, in Trifluoperazine: Clinical and Pharmacological Aspects, Philadelphia, Lea & Febiger, 1958, pp. 47-53.

"... the majority of patients treated—formerly considered practically hopeless—are now in some way more easily managed on the ward."

Klimczynski, J.J.T.: Treatment of Chronically Ill Psychotic Patients with Trifluoperazine: A Preliminary Report, *ibid.*, pp. 101-112.

"The relatively low number of failures of treatment, even in those who have been sick for a long time, must be considered almost as significant as the high proportion of good recoveries."

Goldman, D.: Clinical Experience with Trifluoperazine: Treatment of Psychotic States, *ibid.*, pp. 71-86.

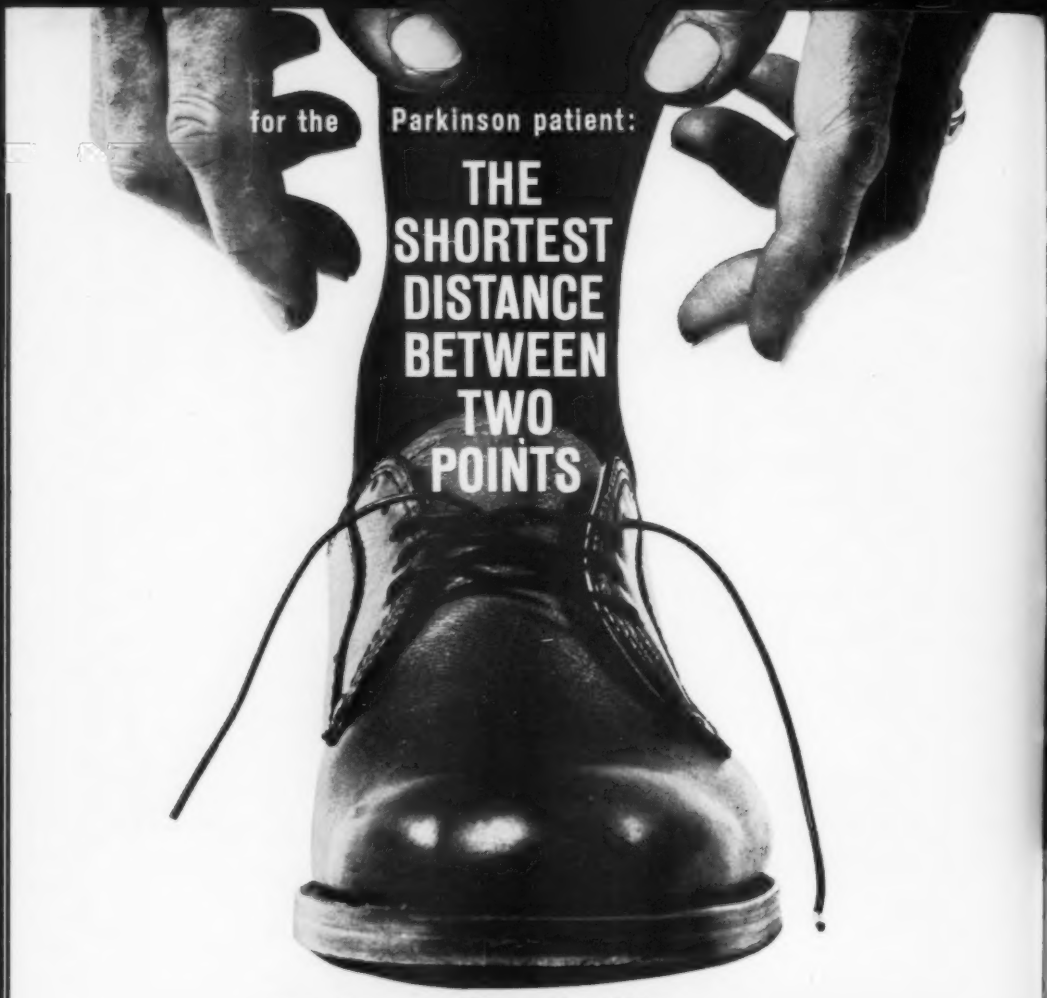
Available: Tablets, 2 mg., 5 mg. and 10 mg.
Multiple dose vials, 10 cc. (2 mg./cc.)

Literature available on request.

Smith Kline & French Laboratories (SKF) **leaders in psychopharmacology**

*Trademark for trifluoperazine, S.K.F.

(10-[3-(1-methyl-4-piperazinyl)-propyl]-2-trifluoromethylphenothiazine)



can be achieved with

ARTANE*

Trihexyphenidyl HCl LEDERLE

Effectively helps restore smooth straight-line function in all forms of Parkinsonism.

One of the best available preparations for sustained control of rigidity and minor tremors.[1] Also active against oculogyria and akinesia.[2] A basic drug for beginning treatment in all types of Parkinsonism.[1, 2]

Continually useful alone or in combination in most cases at any stage. Employable at any age.[1]

Gentle in action at therapeutic dosage.[1] One of the drugs least likely to produce side effects.[3]

Supplied: 2 mg. and 5 mg. TABLETS; ELIXIR, 2 mg. per 5 cc. teaspoonful

Dosage: 1 mg. first day, gradually increased, according to response, to 6–10 mg. daily divided in 3 doses at mealtimes.

1. Doshay, L. J.: M. Clin. North America 40:1401 (Sept.) 1956.
2. Doshay, L. J.: Current M. Dig. 22:11:49 (Nov.) 1955.
3. De Jong, R. N.: J. Michigan M. Soc. 57:722 (May) 1958.

Lederle LEDERLE LABORATORIES, A Division of AMERICAN CYANAMID COMPANY, Pearl River, New York *Reg. U.S. Pat. Of

leads turbulent patients towards rehabilitation

Trilafon

perphenazine

- provides emotional control and social recovery when anxiety, agitation or psychomotor excitement prevail
- keeps patients alert—promotes active cooperation instead of drowsy, passive obedience
- reduces supervisory problems in wards, mess halls and recreation rooms
- offers highest milligram activity of all phenothiazines
- causes no agranulocytosis, significant hypotension or skin rashes and little or no jaundice

TRILAFON Tablets—8 mg., bottles of 50 and 500; 16 mg., bottles of 500.

TRILAFON® REPETABS®—8 mg. (4 mg. in outer layer for *prompt effect* and 4 mg. in inner core for *prolonged action*), bottles of 30 and 100.

TRILAFON Injection—5 mg., ampul of 1 cc., boxes of 6 and 100.

For complete information regarding indications, dosage, side effects, precautions and contraindications consult Schering literature.

Schering

SCHERING CORPORATION • BLOOMFIELD, NEW JERSEY

YR-J-30118

U.S. Pat.

NOW AVAILABLE

more convenient therapy for your myasthenic patients

NEW MESTINON T·I·M·E·S·P·A·N TABLETS

provide, in **slow release form,** the specific antimyasthenic action of Mestinon... smooth, sustained control of symptoms... relative freedom from muscarinic side effects.

The immediate action of a 180-mg Mestinon Timespan is about equal to that of a 60-mg conventional tablet.[1]
The duration of effectiveness of Mestinon Timespan tablets varies from patient to patient, but averages about two-and-one-half times that of regular Mestinon.[2]
Symptomatic control is possible with **fewer daily doses.**

References:
1. K. E. Osserman, **Myasthenia Gravis,** 1958, Grune & Stratton, New York.
2. R. S. Schwab, K. E. Osserman and J. E. Tether, **J.A.M.A., 165**:671, 1957.

MESTINON® BROMIDE—brand of pyridostigmin bromide
TIMESPAN®
ROCHE—Reg. U. S. Pat. Off.

ROCHE LABORATORIES • Division of Hoffmann-La Roche Inc • Nutley 10 • New Jersey

Neurology Volume 9 • Number 1

Dissecting aneurysms of the carotid artery following arteriography

J. F. Ross Fleming, M.D., and Andrew M. Park, M.D.

AMONG THE SERIOUS complications of carotid arteriography is dissecting aneurysm in the wall of the artery at the site of needle puncture, with narrowing or occlusion of the arterial lumen. In this paper, 2 patients are reported in whom this complication has occurred.

Case 1. The first patient was a 58-year-old man with subarachnoid hemorrhage from a berry aneurysm on the right internal carotid artery. After arteriographic study, he developed a dissecting aneurysm of the common carotid artery, with associated hemiplegia. Resection of the involved segment of artery was followed by temporary improvement.

This man had been in good health when, suddenly, he was heard to fall to the floor of the bathroom while he was shaving. On regaining consciousness a few minutes later, he complained of numbness and weakness of his left arm and leg, which lasted about an hour. He also complained of headache and vomiting. When he was examined in hospital a few hours later, no localizing neurologic signs were evident, but there was severe neck rigidity and the spinal fluid was bloody. A course of conservative management was decided upon, and the patient was kept in bed for six weeks. His blood pressure during this time ranged from 160/80 to 130/70.

Six weeks after the initial hemorrhage, a right percutaneous carotid arteriogram was done under local anesthesia. The needle was placed in the common carotid artery close to its bifurcation, and 35% Hypaque solution was used as the contrast medium (Fig. 1). The injection caused considerable pain in the patient's right ear. The internal carotid artery was filled to its bifurcation, and a trace of contrast medium was seen in the middle cerebral artery but none in the anterior cerebral artery. Of particular significance was the presence of a small amount of opaque medium within the wall of the common carotid artery adjacent to the tip of the needle. A small collection of contrast medium near the bifurcation of the internal carotid artery was suggestive but not conclusive evidence of a berry aneurysm.

The next day the patient complained of transient blindness in his right eye and right-sided headache. He had slight weakness of the left side of his face and his left arm, and some sensory inattention was noted in the left hand. Forty hours after the arteriogram, he became drowsy, and during the subsequent three hours he developed a complete left hemiplegia, left homonymous hemianopsia, and impairment of all forms of sensation in the left arm and leg. The right pupil enlarged and ceased reacting to light.

Further arteriographic studies were then done. A needle was placed low in the right common carotid artery (Fig. 2). The contrast medium outlined a large filling defect within the common carotid artery just proximal to its point of bifurcation. The external carotid artery filled, and a small amount of contrast medium within the internal carotid indicated its patency at least up to the base of the skull. Then, a left carotid arteriogram was done (Fig. 3); this showed good filling of the left anterior and middle cerebral arteries and some cross-filling of the right anterior and middle cere-

From the Toronto Western Hospital, Canada.

Read at tenth annual meeting of the Canadian Neurological Society at Toronto, Canada, June 13, 1958.

bral arteries. No aneurysm was demonstrated. A diagnosis was made of thrombosis of the carotid artery following arteriography.

With the aid of hypothermia, the right common, internal, and external carotid arteries were exposed (Fig. 4). A firm lump was palpable in the common carotid artery just below its bifurcation. Pulsations in the internal carotid artery were much more feeble than those in the common carotid. The external carotid was then transected,

Fig. 1. *(Case 1)* Original right carotid arteriogram. Note the collection of contrast medium about the needle tip.

Fig. 2. *(Case 1)* Right carotid arteriogram showing filling defect at bifurcation of common carotid artery impairing the flow in internal carotid

and, on looking into the bifurcation, the lumen was seen to be greatly narrowed; however, the intima appeared intact and no thrombus was obvious within the lumen. The bifurcation was then resected along with the involved portion of the common carotid. The proximal cut end of the internal carotid appeared friable, and intimal atherosclerotic plaques were present. The cut ends of the common and internal carotid arteries were then joined by an end-to-end anastomosis. The circulation had been occluded for a total of thirteen minutes, and the patient's rectal temperature during that time was 29° C.

The operative specimen (Figs. 4 and 5) showed the presence of a large hematoma in the wall of the common carotid artery, raising the intima sufficiently to narrow the lumen from its normal 8-mm. diameter down to a small triangular slit 3 mm. in diameter. This subintimal hematoma was 1.5 cm. long and occupied over half the circumference of the artery. Although its precise relation to the site of needle puncture could not be determined on pathologic examination, it coincided with the area of intramural contrast medium in the initial arteriogram (Fig. 1).

It was evident from the microscopic sections (Fig. 6) that the dissecting aneurysm lay within the layers of the media. Atherosclerotic plaques were present in the common and internal carotid arteries. Of particular interest was the finding of a small mural thrombus on the wall opposite the dissecting aneurysm.

Postoperatively, the patient was more conscious; he talked a little and moved the left arm in response to pain and the left leg spontaneously. His improvement continued for eighteen hours, when he suddenly became deeply comatose and died within a few hours.

At autopsy there was marked swelling and softening of the right cerebral hemisphere. A small berry aneurysm, containing a recent thrombus, was present at the bifurcation of the right internal carotid artery. The intracranial portion of the internal carotid artery and its branches were all patent. The lumen of the right common carotid artery (Fig. 7) was obliterated by an extensive dissecting aneurysm that extended from the site of anastomosis down to the arch of the aorta; this presumably arose from the arterial suture line.

Case 2. The second patient was a woman who died from a massive intracerebral hematoma. A percutaneous carotid arteriogram had been done twelve hours before death, and, at autopsy, a small dissecting aneurysm of the common carotid artery was an incidental finding.

The patient was 32 years of age, with a previous record of good health. She was four and a half months pregnant. Four days before death, she complained of headache, stiff neck, and vomiting. Within the next eighteen hours, she developed left hemiplegia and, on the day before death, a dilated right pupil. A right percutaneous carotid arteriogram (Fig. 8) showed elevation of the mid-

dle cerebral artery indicating the presence of a massive intratemporal lesion; however, a possible source of hemorrhage was not evident from the arteriogram. There had been some difficulty cannulating the artery, and it is certain that the artery was punctured several times. The needle tip could be seen in contact with the posterior wall of the artery, and, after the contrast medium had left the arterial lumen, a small amount of it remained in the layers of the arterial wall. At the time of arteriography, her blood pressure was 130/80, though it subsequently rose to much higher levels. A large hematoma was removed from deep in the right cerebral hemisphere, but the patient died a few hours later.

At autopsy, the cause of death was found to be severe cerebral swelling, associated with temporal and cerebellar pressure cones. An incidental finding was a dissecting aneurysm 6 cm. long in the wall of the right common carotid artery (Fig. 9). It extended from 5 cm. above the aortic arch to just below the bifurcation of the common carotid artery. The dissection consisted of a thin column of blood, 1 to 2 mm. thick, completely surrounding the vessel and lying within the layers of the media of the artery. The lumen of the vessel was slightly narrowed. There were no thrombi within the lumen and no appreciable atherosclerosis of the vessel wall. Near the upper end of the dissection were 4 small linear puncture wounds of the intima. Microscopic sections of this area (Figs. 10 and 11) showed communication of the needle tract with the hemorrhage in the arterial wall. Elastic stains verified that the hemorrhage lay within the media.

The patient's normal blood pressure was not known, but the finding of arteriolar nephrosclerosis at autopsy suggested previous hypertension, perhaps aggravated by pregnancy. Furthermore, the possibility of an abnormal bleeding tendency was suggested by the presence of large ecchymoses of the trunk and extremities, together with a history of frequent bruising. However, no premortem tests were done to confirm this.

DISCUSSION

That local complications may result from a percutaneous carotid arteriogram is well known. Such lesions as thrombus formation at the puncture site, the formation of false aneurysms, carotid-jugular fistulas, and the creation of a flap of intima which occludes the lumen as a valve have all been described.

There are a few reports on record of dissecting aneurysm as a complication of carotid arteriography. In most instances, the hemorrhage was small and asymptomatic as in our second patient. Large lesions such as occurred in our first patient have been surprisingly uncommon. Rimpau[1] from Hamburg studied the

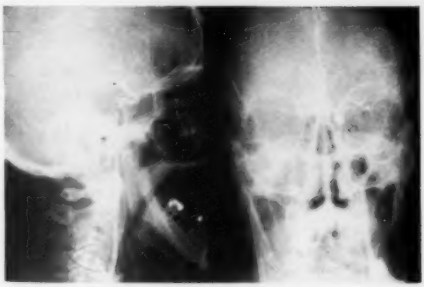


Fig. 3. *(Case 1)* Left carotid arteriogram

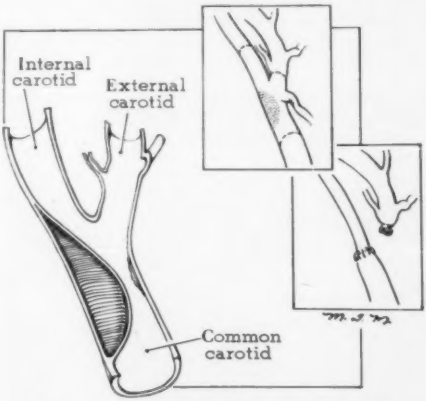


Fig. 4. *(Case 1)* Sketch of operative findings. Inset is shown area of resection and anastomosis.

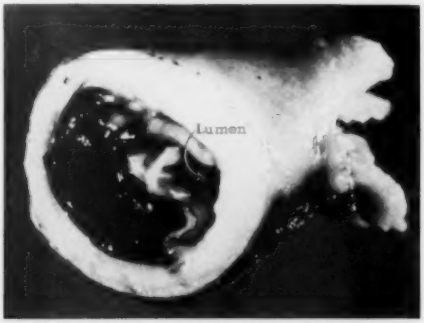


Fig. 5. *(Case 1)* Photograph of operative specimen, looking into cut end of common carotid artery. The lumen is greatly narrowed by the large blood clot deep to the intima.

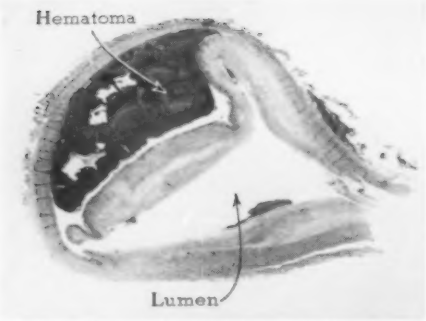


Fig. 6. *(Case 1)* Photomicrograph of operative specimen. Note narrowing of lumen by large subintimal blood clot and small intimal thrombus lying on opposite wall of artery.

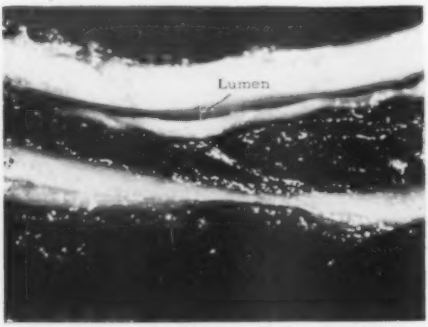


Fig. 7. *(Case 1)* Photograph of carotid artery at autopsy. The lumen is the narrow slit in upper portion of the picture, and the large dissecting aneurysm lies within the layers of the media. The origin of this lesion was not the arteriogram but probably the surgical anastomosis.

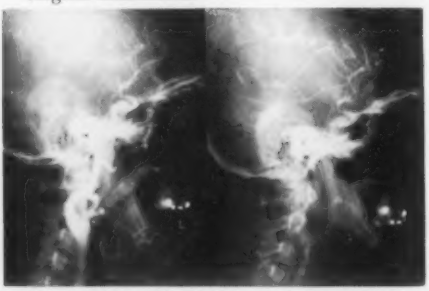


Fig. 8. *(Case 2)* Right carotid arteriogram showing elevation of middle cerebral artery. Note contrast medium remaining in the wall of the carotid artery in picture on the right.

autopsy findings in 92 patients who had had percutaneous carotid arteriograms done; intramural hematomas at the puncture site were found in 11. In 4 of these, the lesion was pea size; in 1 case, a patient with periarteritis nodosa, it was large enough to narrow the lumen severely. In another patient, the hemorrhage had dissected down to the aorta. In a later review, Rimpau[2] reported 3 instances of complete occlusion of the artery by a hematoma within the wall. Perhaps Rimpau's most significant finding was the presence of small lacerations of the intima on the wall opposite the puncture site in nearly every patient who had had an arteriogram, indicating that the artery was almost always transfixed by the needle. This posterior wall puncture wound varied from 1 to 3 mm. in length. In 1 patient, a laceration of the intima extended around one-half the circumference of the artery; the distal edge of the intima was pushed out into the blood stream and the flap so formed acted as a valve. Sirois[3] has reported a similar case. These lacerations of the posterior wall intima are probably associated with the production of dissecting aneurysms, as will be discussed below.

DeGrood[4] in Holland reported a case of occlusion of the common carotid artery caused by dissecting aneurysm which occurred during arteriography. The films showed a complete block in the common carotid artery, and contrast medium was seen in the axillary artery. During the injection, the patient had experienced pain in his neck, shoulder, and arm but not in his face. As the diagnosis of carotid artery occlusion did not fit the clinical picture, an air study was done, revealing a frontal lobe glioma which was removed. The patient died, and, at autopsy, a dissecting aneurysm in the common carotid artery 7 cm. long was seen.

Rowbotham[5] reported a case of hemorrhage in the media 1 cm. long and 0.5 cm. thick, and he also noted the presence of puncture wounds of the posterior wall intima in all carotid arteries studied. Crawford[6] at St. George's Hospital, London, reviewed 75 autopsy patients who had had carotid arteriography. He found 9 patients with small dissecting aneurysms 0.5 to 4 cm. long. It is significant that in the large autopsy series of

both Crawford and Rimpau, small dissecting aneurysms were found in over 10% of all patients submitted to carotid arteriography. However, it is reassuring that hematomas large enough to narrow the lumen severely and thereby cause symptoms are extraordinarily rare.

It is of interest that a case similar to ours recently prompted Drs. W. A. Warner and A. Earl Walker[7] to attempt reproduction of this condition in dogs. They apparently were unsuccessful in producing a dissecting aneurysm in a dog's carotid artery even by very vigorous attempts at needling and injecting contrast medium.

A dissecting aneurysm may be produced at either [1] the *puncture site* or [2] the *site of perforation of the intima of the posterior artery wall.*

1] Aneurysm would be likely to occur at the puncture site (Fig. 12) if a dull or barbed needle were used. This might tear apart the layers of the artery as it penetrated and create a space for subsequent accumulation of hemorrhage, the blood entering this space from the arterial lumen via the needle tract after the needle is withdrawn. It follows that the needle should be perfectly sharp and as small in diameter as feasible.

2] It is more probable that dissecting aneurysms arise from the posterior wall of the artery at the site of transfixion by the needle (Fig. 13). Presumably, the bevel of the needle is partly within the lumen of the artery and partly subintimal. The injection of contrast medium then dissects up the intima, as seen in the roentgenograms of both our cases (Figs. 1 and 8), leaving a space for subsequent accumulation of hemorrhage. As it is usually impossible to avoid transfixing the artery during puncture, the needle tip should be withdrawn from the opposite wall and then threaded up the lumen. The use of a short beveled needle should lessen the likelihood of intramural injection.

Considering the rarity of postarteriographic dissecting aneurysms in man and Warner and Walker's inability to produce this condition in dogs, it seems likely that other factors in addition to these mechanical ones must play a part in predisposing to the formation of this lesion. An artery wall made friable by atheroma, an

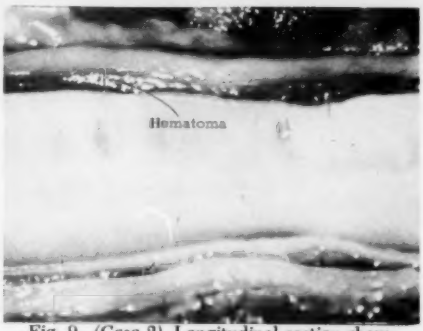


Fig. 9. *(Case 2)* Longitudinal section showing interior of right common carotid artery at autopsy. The dissecting aneurysm appears as a thin layer of blood just outside the intima. Note particularly several needle puncture marks on the intima.

Fig. 10. *(Case 2)* Photomicrograph of longitudinal section of posterior wall of right common carotid artery. Two distinct needle tracks are marked by arrows, and one of these goes well into the media. The large dissecting aneurysm is seen deep to the intima.

Fig. 11. Higher magnification of Fig. 10, showing needle tracks.

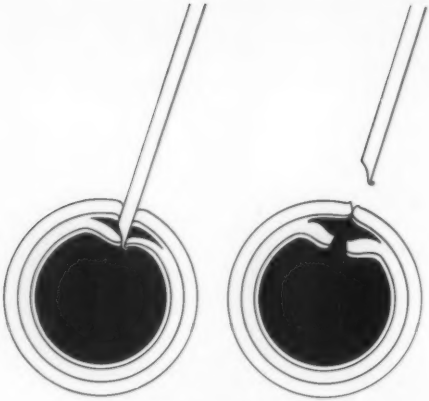


Fig. 12. Possible method of production of dissecting aneurysm after use of barbed needle

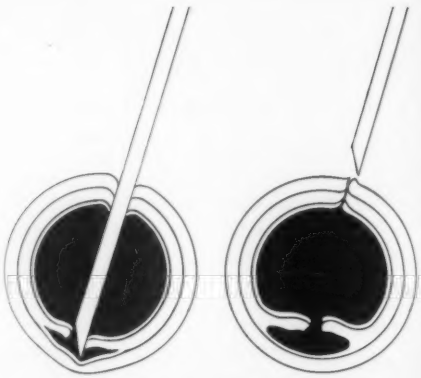


Fig. 13. Probable method of production of dissecting aneurysm by puncture of posterior wall of artery

elevated blood pressure, or an abnormal bleeding tendency may be important in this regard. The increased risk of subjecting patients with arterial disease to arteriography should be fully realized.

SUMMARY AND CONCLUSIONS

1] Occurrence of dissecting aneurysm of the common carotid artery after arteriography in 2 patients is reported. In 1 patient, the artery was narrowed sufficiently to produce massive cerebral infarction. In the other, the hemorrhage was small and was an incidental autopsy finding.

2] It is surprising how resistant the vessel wall is to the trauma of puncture. However, large autopsy surveys indicate that a significant percentage of patients subjected to arteriography develop a small dissecting aneurysm. Lesions large enough to occlude the lumen are extraordinarily rare, but a few have been reported. Nevertheless, even the smallest intimal elevation may become the site of thrombus formation and, thus, a possible source of emboli.

3] The clinician should be alert to this complication of arteriography, which may explain certain postangiographic symptoms and may elucidate the interpretation of incomplete filling of the carotid artery in some cases.

REFERENCES

1. Rimpau, Arnold: Zur Morphologie der Carotispunktion. Virchows Arch. 330:156, 1957.
2. Rimpau, A., and Seils, H.: Pathologisch-anatomische Befunde an der Punktionsstelle bei der Hirnarteriographie und Betrachtungen zur Punktionstechnik. Fortsch. Röntgenstrahl. 87:191, 1957.
3. Sirois, J., LaPointe, H., and Cote, P.: Unusual local complication of percutaneous cerebral angiography. J. Neurosurg. 11:112, 1954.
4. DeGrood, M. P.: Dissecting aneurysm of carotid following arteriography. Rev. Neurol. 90:661, 1954.
5. Rowbotham, G. F., and others: Technique and dangers of cerebral angiography. J. Neurosurg. 10:602, 1953.
6. Crawford, T.: Pathological effects of cerebral arteriography. J. Neurol. Neurosurg. & Psychiat. 19: 217, 1956.
7. Warner, W. A., and Walker, A. Earl: Discussion of paper at Tenth Annual Meeting of Canadian Neurological Society at Toronto, Canada, June 13, 1958.

Permeability of cerebral blood vessels in healing of brain wounds

J. Lee and J. Olszewski

ROZDILSKY AND OLSZEWSKI have shown that, under certain conditions, the cerebral blood vessels become permeable to the protein molecules.[1] For example, increased permeability was observed in an area surrounding an acute brain wound produced by heat coagulation. The purpose of experiments reported here was to study how long such an increased permeability would last and how its disappearance correlated with the healing of the wound. In addition, we wanted to know whether the axonal degeneration along the pathways leading from or to the damaged cortex and the retrograde degeneration developing in the thalamic nuclei would be accompanied by increased permeability.

MATERIAL AND METHOD

Experiments were performed on rabbits. Under Nembutal anesthesia, a midline incision of the scalp was made; then an area of parietal bone was cleaned from the periosteum on one side of the midline over a surface of approximately 2 sq. cm. A brass rod, 0.3 cm. in diameter, was heated over the gas flame for three minutes, and the end of the rod pressed firmly to the bone for twenty seconds. (The rod was applied for ten seconds in Experiment 104.) This procedure was repeated four times. The rod was applied each time to a new region. Then, the skin was sutured, and the animal allowed to recover from anesthesia. With this technic, we were able to produce a cortical necrosis of approximately 1 cc. in diameter extending to, or slightly encroaching on, the underlying white matter.

To test the changes of permeability, iodinated, radioactive bovine albumin was used. At various time intervals after the cortical coagulations – from a few minutes to thirty-seven days – intravenous injections of radioactive albumin in doses of 1 mc. were given. The amount of solution injected varied from 3.5 to 5.5 cc. The concentration of albumin in the solution was 2% in all experiments. The iodinated, radioactive bovine albumin was obtained commercially from Charles Frosst, Montreal, and, according to the manufacturer, did not contain more than 2% of unbound iodine.

An exchange perfusion was begun two hours after the injection of albumin. Perfusion consisted of an intravenous drip of 6% Dextran in saline* into the marginal vein of one ear and simultaneous bleeding from the artery of the other ear. The drip and the bleeding were maintained at approximately the same rate of 5 cc. a minute. After a half hour of perfusion, the blood was visibly diluted, and the animal became somnolent and, then, unresponsive. Respiration was fast and labored. Death occurred after approximately sixty minutes of perfusion; 300 cc. of Dextran and of blood escaped. When respiration and heart beat stopped, the animal's head was perfused with 100 cc. of saline through both common carotid arteries, followed by 200 cc. of 30% alcohol in 10% formalin. The brain was removed either immediately or after two hours. One or two thin coronal slides which included the brain wound were made and embedded overnight in paraffin. The rest of the brain was placed in alcohol-formalin solution for further fixation. The next morning, paraffin sections 25 and 15 μ thick and frozen sections 50 μ thick were prepared. After drying, these sections

From the Laboratory of Experimental Neuropathology, Department of Pathology, University of Saskatchewan, Saskatoon, Sask.
Supported by a grant from the Canadian National Research Council.

*The assistance of Abbott Laboratories, Ltd., in donating Dextran for this study is acknowledged.

were placed in contact with No-Screen x-ray films. The autoradiograph was developed after four days. The histologic sections were stained by Nissl, hematoxylin-eosin, and periodic acid-Schiff methods.

This type of exchange perfusion was used to remove all blood from the vessels. Obviously, the presence of blood in the vessels would obscure the autoradiographic pattern by causing darkened emulsion proportional to the vascularity of a given region.

RESULTS

With the method of perfusion described, no radioactivity of the healthy meninges and brain was observed – with the exception of the hypothalamus and choroid plexus. In none of the experiments was there any radioactivity in the thalamus or along the long efferent or afferent tracts. In several of the rabbits, a so-called spontaneous rabbit encephalitis was an accidental finding. The lesions consisted of perivascular cuffing by lymphocytes and of small granulomas formed by epithelioid cells surrounded by lymphocytes and often having necrotic centers. In none of these animals was there any radioactivity in relation to these lesions. The pathologic lesions resulting from coagulations and the corresponding autoradiographic pattern are described under individual experiments.

EXPERIMENT 138

The tagged albumin was injected immediately after cortical coagulation.

Pathology. In the necrotic area, the nerve and glia cells partly lost their stainability, many small hemorrhages were present, and the tissue was edematous, merging gradually into healthy surrounding tissue. No inflammatory cells were seen. No change occurred in the white or gray matter farther away from the lesion.

Autoradiograph. Intense radioactivity occurred in and around the area coagulated (Fig. 1). The radioactivity spread to the white matter along the lateral wall of the lateral ventricle reaching the hippocampal formation. It also spread subpially in the superficial cortical layers in the same direction, but for a shorter distance. In addition, radioactivity extended along the corpus callosum to the opposite hemisphere and was present in the hippocampal formation where this was near the area coagulated. A narrow rim of radioactivity was also present over the free surface of the whole brain and over the ventricular surface, causing a thin line which marked the border of the tissue.

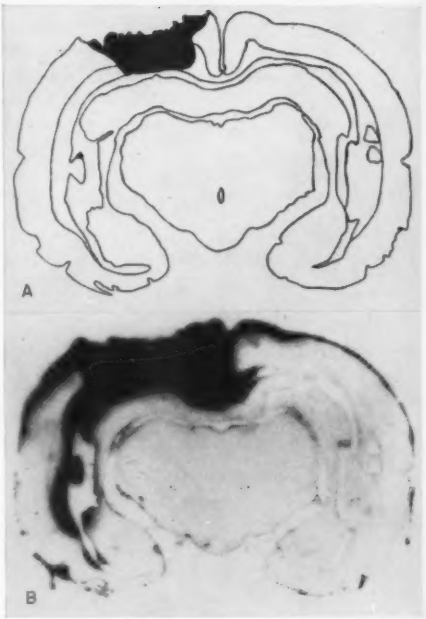


Fig. 1. Experiment 138. *(A)* Diagram of the coronal section of the brain with coagulation necrosis marked black. *(B)* The corresponding autoradiograph

EXPERIMENT 139

The tagged albumin was injected two hours after coagulation. The pathologic changes and the extent of radioactivity were the same as Experiment 138.

EXPERIMENT 141

The tagged albumin was injected twenty-eight hours after the coagulation.

Pathology. The necrotic region was edematous and hemorrhagic, and the extravasated blood partly hemolyzed. The nerve and glia cells were stained very lightly. The polymorphonuclear cells were abundant in the overlying meninges and on the border of the

dead tissue. No difference was found in the appearance of the white matter of the left and right hemispheres.

Autoradiograph. Radioactivity was present over an area smaller than that of Experiments 138 and 139. Radioactivity was not seen in the opposite hemisphere and extended laterally in the white matter for considerably shorter distance (Fig. 2). The subpial extension was also shorter. The central portion of the necrotic tissue was less radioactive than the periphery. Hardly any radioactivity was present either along the pia or the ependyma farther away from the lesion.

EXPERIMENT 148

The tagged albumin was injected four days after the coagulation.

Pathology. In the necrotic region, the stainability of the nerve and glia cells was almost lost. Numerous pools of hemolyzed red blood cells were present. The region was surrounded by a dense wall of polymorphonuclear cells in various stages of nuclear disintegration. Outside this wall, the tissue was edematous and contained a few small hemorrhages and a moderate amount of histiocytes. At the border of necrosis, young fibroblasts were streaming from the meninges into the cortex – wedging themselves between dead and healthy tissue.

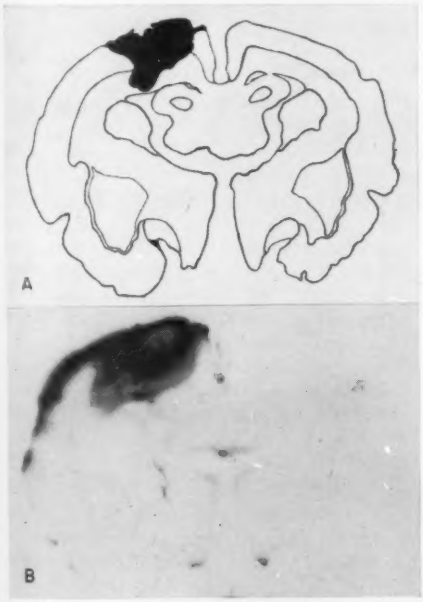


Fig. 2. Experiment 141. *(A)* Diagram of the coronal section of the brain with necrotic region marked black. *(B)* The corresponding autoradiograph

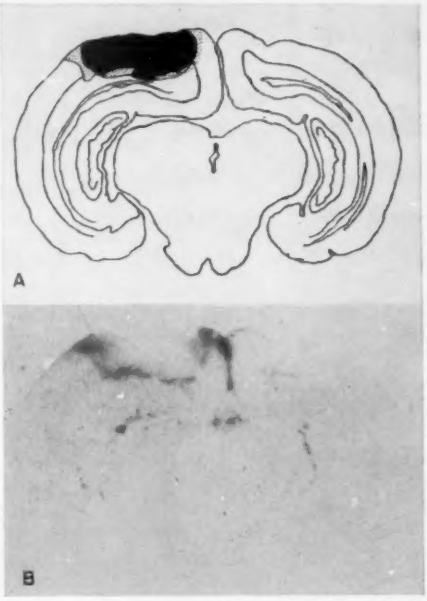


Fig. 3. Experiment 148. *(A)* Diagram of the coronal section of the brain with necrotic region marked black. Areas of fibroblastic proliferation punctuated. *(B)* The corresponding autoradiograph

Autoradiograph. Moderate radioactivity was present at the border of the necrotic region and in the meninges laterally and medially to the necrosis (Fig. 3). The center of the necrotic area was not radioactive. The radioactivity surrounding it corresponded to the edematous tissue and to the regions with active fibroblastic proliferation.

EXPERIMENT 104

The tagged albumin was injected nine days after coagulation.

Pathology. In this experiment, which was conducted in an earlier stage of the investi-

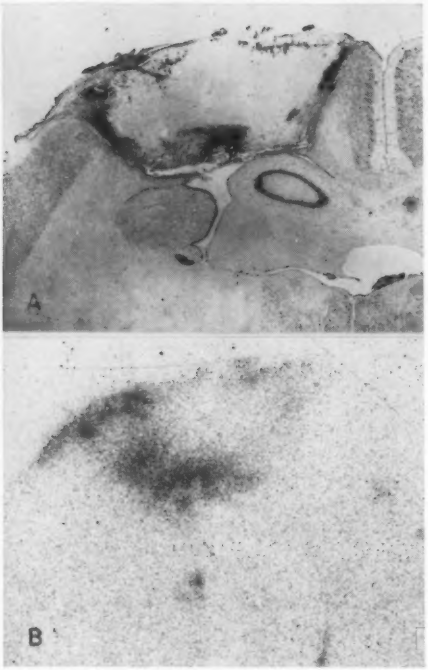


Fig. 4. Experiment 140. *(A)* Low-power photomicrograph of the Nissl-stained section showing details of the lesion. *(B)* The corresponding autoradiograph

gation, the heated rod was applied each time for ten seconds only and the resulting coagulation involved only the first, second, and a part of the third cortical layers. No necrotic tissue was present, and the cortical defect was filled by granulation tissue continuous with the thickened meninges. Proliferating fibroblasts, macrophages, and lymphocytes formed this granulation tissue.

Autoradiograph. Moderate radioactivity occurred in the granulation tissue with very faint extension for about 2 mm. into the underlying cortex.

EXPERIMENT 140

The tagged albumin was injected fourteen days after coagulation.

Pathology. The lesion was similar to that of Experiment 148, but the organization was in a more advanced stage. The meninges over the lesion were thickened and contained many young fibroblasts and macrophages. The fibroblasts streamed from the meninges around the lesion, walling it off from the surrounding cortex. Fragmented nuclear debris was present in small patches inside of the fibroblastic layer, and numerous macrophages lay among the fibroblasts. The center of the lesion completely lost its stainability with the Nissl method. The edema around the lesion was slight.

Autoradiograph. A moderate radioactivity was present in the border of the lesion and in the overlying meninges. The center of the necrosis was not radioactive. Only slight extension of the radioactivity along the meninges was seen (Fig. 4).

EXPERIMENT 143

The tagged albumin was injected twenty-one days after the cortical coagulation.

Pathologic changes and autoradiographic pattern were comparable to those in Experiment 140. Granulation tissue separating the necrotic area from the surrounding tissue was the most radioactive.

EXPERIMENT 137

The tagged albumin was injected twenty-eight days after cortical coagulation.

Pathology. The coagulated area was filled by densely packed macrophages – some of them containing blood pigment. There were very few fibroblasts. The meninges over the coagulated area were not on the section and could not be assessed. Slight glial proliferation was present in the tissue beneath the lesion.

Autoradiograph. The surface of the brain in the region of coagulation was slightly darkened.

EXPERIMENT 144

Tagged albumin was injected thirty-seven days after cortical coagulation.

Pathology. Appearance of the coagulated area was the same as in Experiment 137. In these sections, the meninges were present on the coagulated cortex. They were moderately thickened and contained fibroblasts and macrophages.

Autoradiograph. Slight darkening corresponded to the young fibroblastic tissue of the meninges overlying the lesion (Fig. 5).

DISCUSSION

1] Radioactivity in the degenerating white matter and in the thalamic nuclei: No darkening of the photographic emulsion was observed over regions which were undergoing retrograde or Wallerian degeneration, indicating that the vascular impermeability for large molecules is a stable characteristic which does not change under these conditions.

2] Radioactivity of the necrotic tissue: The necrotic tissue showed intense radioactivity if the tagged albumin was injected immediately or two hours after the coagulation. At this stage, gross damage to the blood vessels must have occurred, with outpouring of plasma and cellular elements into the tissue. This process was not operating longer than twenty-four hours, since, twenty-eight hours after coagulation, the necrotic mass was less radioactive in its center than at its border. Massive extravasation of blood was not taking place, and the necrotic area was undergoing gradual organization.

3] Radioactivity in the immediate vicinity of the necrotic region: At the earliest stages after coagulation, the intense radioactivity of the whole region obscured any details that might have been present at the border of the lesion. From the twenty-eighth hour, moderate radioactivity surrounded the necrotic region and localized in immediately adjacent tissue. Because of the poor resolution of autoradiographs on roentgenograms, the site of greatest radioactivity could not be localized precisely. We had the impression, however, that it was concentrated in the edematous tissue surrounding obvious necrosis and also in the regions of fibroblastic proliferation. This latter observation was supported by results obtained when albumin was injected thirty-seven days after coagulation. Minimal radioactivity was present only over the surface of the brain and corresponded to the thickened meninges, whereas underlying accumulation of histiocytes was not radioactive.

Possibly, radioactivity of fibroblastic tissue should be compared not with that of the brain, but with radioactivity of organs in which permeability of capillaries is not characterized by the presence of a specific barrier. We prepared autoradiographs of the tongue, the parotid gland, and the anterior lobe of the pituitary from a healthy animal injected with tagged albumin and perfused. These autoradiographs showed moderate radioactivity. This observation agrees with a now well-established fact – vessels of most organs are permeable to serum proteins. Hence, we suggest that the radioactivity of the young fibroblastic tissue reflects a characteristic proper to most other organs, but not to the brain, and may have nothing to do with the inflammatory origin of this tissue. Apparently, a significant difference is found between the brain and the other organs in relation to radioactivity from intravenous injection of heterologous, iodinated albumin. We may add that the brain itself is not entirely uniform in this respect, since the tuber cinereum and the choroid plexus were always slightly radioactive. We have not prepared sections through the area postrema, but expect that it, too, would be radioactive. These are the same regions which under normal conditions are permeable to trypan blue, and it is interesting that, under normal conditions, the

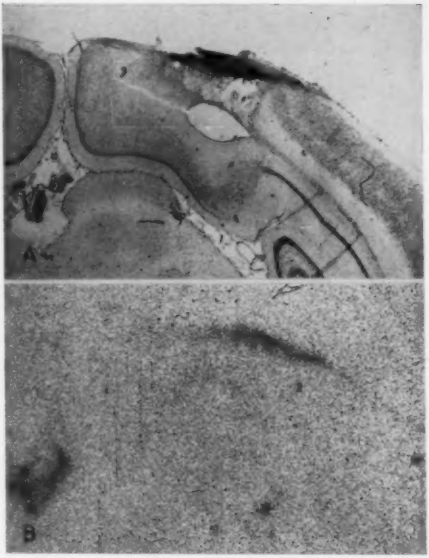


Fig. 5. Experiment 144. *(A)* Low-power photomicrograph of a Nissl-stained section showing details of lesion. *(B)* The corresponding autoradiograph. Darkening in left lower corner overlies the choroid plexus.

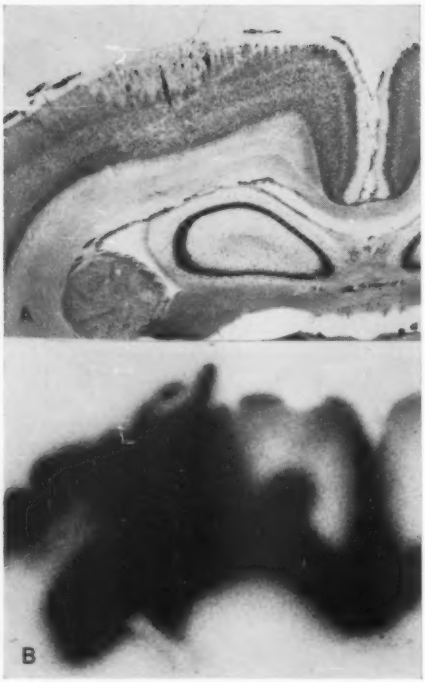


Fig. 6. Experiment 138. *(A)* A low-power photomicrograph of a Nissl-stained section through the periphery of the coagulated region. The lesion occupies only half the thickness of the cortex. The surrounding tissue is unchanged. *(B)* The corresponding autoradiograph. The darkened emulsion extends beyond the necrosis. The cortex on both sides of the lesion is spared.

permeability to 2 different substances or molecular sizes is so similar.

4] Radioactivity in regions distant from the necrosis: If tagged albumin was injected immediately after coagulation or two hours later, slight radioactivity was observed all along the pia-arachnoid and ependyma. In these early stages after coagulation, the blood vessels are damaged enough to allow gross passage of the plasma to the cerebrospinal fluid. From there, the radioactive iodine either unbound or coupled with albumin may enter the most superficial subpial or subependymal tissue. Substances which otherwise do not pass the blood-brain barrier can enter the brain via cerebrospinal fluid, according to other investigators.[2–4] We have confirmed this for albumin in as yet unpublished experiments.

If tagged albumin was injected within twenty-eight hours after coagulation, much more intense radioactivity was present around the lesion in the white matter and in the superficial cortical layers. Radioactivity was not present if the injection was made four days or longer after injury. The distribution of this radioactivity was characteristic because it seemed to have the tendency to spare the gray matter and to spread in the white matter (Fig. 6). Thus, the dark area of the photographic emulsion was not extending the same distance in all directions; the dark area reached farther over the white matter and the subpial cortical layers than over the cortical layers.

The intense radioactivity in the superficial cortical layers surrounding the lesion is probably the result of penetration of the radioactive, extravasated serum from the subarachnoid space into the brain tissue. We have mentioned that we obtained experimental evidence for the existence of such a mechanism. As could be expected, the radioactivity was more intense close to the lesion and faded away peripherally.

The relative sparing of the gray matter and the more intense radioactivity in the white matter can be explained by two hypotheses. [A] The extravasated radioactive serum may spread along the nerve fibers of the white matter more easily than in the gray matter. [B] The radioactivity may result from the increased permeability of the vessels at a distance from the lesion.

In favor of the first hypothesis is the observation from human neuropathology that the cerebral hemorrhage often dissects the white matter spreading along the fiber tract.

To accept the second hypothesis, we must assume a special vulnerability of the vessels of the white matter as compared with those of the gray matter. It is well known that gray matter is more vascular and, if all vessels were equally damaged, the gray, and not the white matter, should have been more radioactive. Such is the case if vessels are damaged directly by toxic substances. In as yet unpublished experiments, Lee has injected 30% ethanol into the common carotid artery of rabbits followed

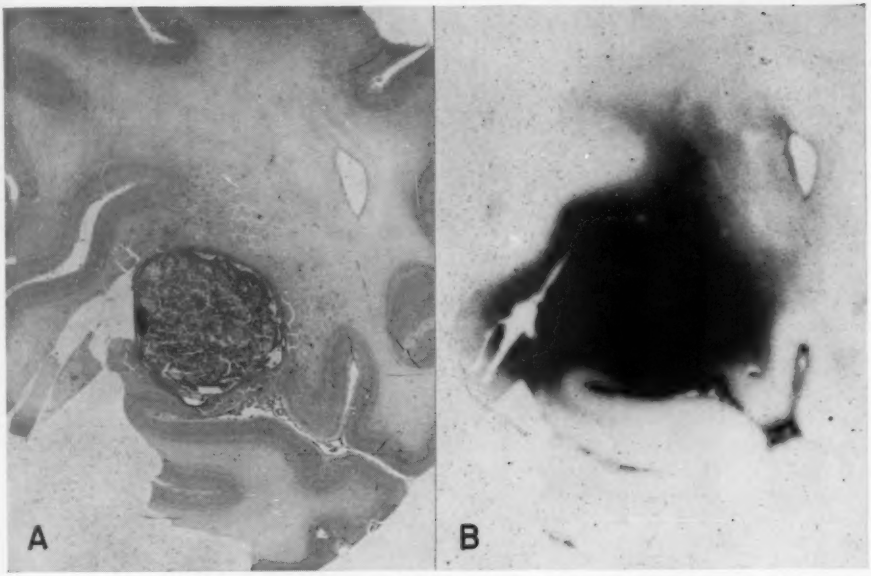


Fig. 7. *(A)* Histologic section of the occipital lobe showing a large metastatic tumor. *(B)* The corresponding autoradiograph. The radioactivity affects the white matter but spares the cortex.

by intravenous injection of iodinated albumin. Autoradiographs made from the brains of these animals showed much more intense darkening over the gray than over the white matter. It should be mentioned that brain edema is known to affect the white matter more intensely than the gray matter and thus parallels the autoradiographic effect seen in our experiments. We observed a striking example of this in a patient with metastatic thyroid carcinoma of the brain. This patient was administered 100 mc. of radioactive iodine four days before death. The neurohistologic investigation revealed that the tumor was surrounded by necrotic and edematous tissue. The autoradiograph (Fig. 7) showed the darkening of the emulsion which was not circular around the neoplasm, but followed the white-gray matter border, sparing the cortex.

We do not intend to indicate that the lesion of the blood vessels is the only, or even the main cause, of cerebral edema in all instances. Cerebral edema can be produced by 3 main mechanisms: *(a)* changes in the osmotic pressure of the blood, *(b)* changes in the osmotic pressure of the tissue, and *(c)* increased permeability of the blood vessels. In our experiments, the mechanism *(c)* must have been in operation, since, in the nervous system, the leakage of proteins from the blood into the tissue has to be accompanied by altered vascular permeability. Thus, we propose as a working hypothesis that cerebral edema localized mainly in the white matter may be due primarily – or predominantly – to the alteration of vascular permeability. We can speculate further that the cerebral edema due to either mechanism *(a)* or *(b)* may show a different site of predilection. Further experimental work is obviously needed on these problems.

The last point we would like to discuss is the relationship between the histologic appearance of the white matter and its radioactivity. It is noteworthy that in the experiments in which tagged albumin was injected during the first two hours after cortical coagulation, the appearance of the radioactive white matter did not differ from that of the nonradioactive matter. However, in other experiments not yet published, we were able to

demonstrate edema of the white matter if animals were killed sixteen or twenty-four hours after cortical coagulation. This edema was most pronounced in the subependymal white matter and thus corresponded to the region which was intensely radioactive in the present series of experiments. These results are in accordance with findings of Klatzo,[5] who, in experiments on cats, observed edema in the white matter if the animals were sacrificed from ten to twenty-four hours after the lesion was made. Possibly, submicroscopic changes of tissue may precede the histologic evidence of edema by several hours.

SUMMARY

1] Experiments were performed on rabbits to study changes of vascular permeability while cerebral wounds healed and during retrograde and Wallerian degeneration. Radioactive, iodinated bovine albumin and autoradiographic methods were used to assess permeability changes.

2] Under normal conditions, cerebral blood vessels are impermeable to albumin – with the exception of such regions as the hypothalamus or the choroid plexus which are known to have vascular permeability different from that of most parts of the brain.

3] No increased permeability for albumin molecules was seen in regions undergoing retrograde or Wallerian degeneration.

4] Heat coagulation of a small region of the frontoparietal cortex produced increased permeability, according to the radioactivity of the tissue.

5] In earliest stages after coagulation, radioactivity was very intense and corresponded to the necrotic region and the surrounding tissue. In the surrounding tissue radioactivity spread farther in the white matter than in the gray matter. Distribution was similar to that of brain edema.

6] In the later stages (fourth and twentieth day), the central cores of the dead tissue and surrounding healthy tissue were becoming progressively less radioactive. The radioactivity was finally confined to the border of the necrosis and to the overlying meninges.

7] In the last stages investigated (twenty-eighth and thirty-seventh day), the radioactivity was confined to the thickened meninges overlying the lesion. It was considered that this radioactivity was a manifestation of the usual property of the connective tissue.

REFERENCES

1. Rozdilsky, B., and Olszewski, J.: Permeability of cerebral blood vessels studied by radioactive iodinated bovine albumen. Neurology 7:270-279, 1957.
2. Goldmann, E. E.: Vitalfarbund am Zentralnervensystem. Berlin, Eimer, 1913.
3. Bakay, L.: Blood-brain barrier; with special regard to the use of radioactive isotopes. Springfield, Ill., Charles C Thomas, 1956.
4. Koenig, H.: Production of injury to the central nervous system with a nucleic acid antimetabolite. Anat. Rec. 130:326-327, 1958.
5. Klatzo, I.: Personal communication.

Role of gamma-aminobutyric acid in electrocortical arousal

E. Eidelberg, M.D.,* S. Feldman, M.D.,† and
H. W. Magoun, Ph.D.

IT HAS RECENTLY been proposed that electrocortical arousal — as manifested by a change of high voltage slow activity to low voltage fast activity in the electroencephalogram — involves the generation of inhibitory postsynaptic potentials in the apical dendrites of the cortex by an enhanced activity of cortical inhibitory interneurones.[1,2] This theory has been based on the often demonstrated fact that arousal induced by peripheral or electrical stimulation of the brain stem reticular formation interferes with recruiting, augmenting, and primary auditory and direct cortical responses.[1–6]

A good deal of evidence has been presented lately to show that gamma-aminobutyric acid (GABA), normally present in mammalian gray matter, has effects on cortical evoked potentials compatible with an enhanced inhibitory activity, be it as a blocking agent of excitatory synapses[7,8] or as an inhibitory synaptic transmitter.[9–11] We, therefore, decided to test the effects of its addition by topical application and its depletion by administration of thiosemicarbazide[12–14] on these arousal effects on evoked potentials.

METHODS

Unanesthetized adult rabbits were employed exclusively. After necessary surgical procedures had been carried out under light etherization and all painful points infiltrated with procaine, the animals were immobilized with gallamine triethiodide (Flaxedil) and kept under artificial ventilation. Under these conditions, a quite stable "sleep" electroencephalographic record can be obtained without need of central anesthesia or brain stem lesions; electroencephalographic "arousal" can be obtained by peripheral or reticular stimulation.

To minimize the possibility of interference by current spread into long ascending pathways, arousal was always obtained by electrical tetanic stimuli delivered to the dorsal midbrain reticular formation at the P_8 plane of Sawyer, Everett and Green's Atlas.[15]

Stimulation was delivered through concentric pairs of stainless steel electrodes by means of Grass S4C stimulators with their isolation units. Recording from the cortex was carried out by chlorided silver ball electrodes, monopolarly with respect to an indifferent reference electrode in the wound margin. Grass P_5 R-C coupled amplifiers, a DuMont 333 double beam CRO, and a Grass camera were used to record individual responses and a Grass III-D 8-channel inkwriting oscillograph to monitor the cortical electrical activity.

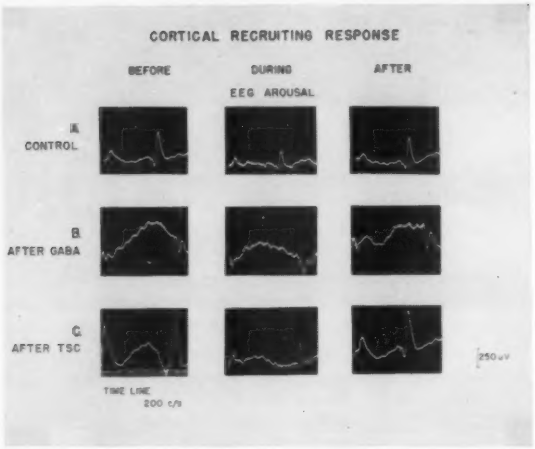
In many of the experiments, the stimulation was monitored through an additional oscilloscope system in which the current delivered per shock could be read directly from the tube face.

GABA was applied topically on the cortex as a 1% aqueous solution with a *p*H of 7.2. Thiosemicarbazide was administered intravenously at a dosage of 20 mg. per kilogram or applied topically on the cortex as a 0.5% buffered solution. All deep electrode placements were checked on serial frozen sections stained with cresyl violet.

RESULTS

The effects of topically applied solutions of GABA to the cortex on both spontaneous and evoked cortical activities have been described

From the School of Medicine, University of California at Los Angeles, and the Veterans Administration Hospital, Long Beach, Calif.
*Lima, Peru
†Hadassah University Hospital, Jerusalem, Israel
Supported by a grant (B-611) from the National Institute of Neurological Diseases and Blindness, United States Public Health Service.



Paired shocks to centrum medianum, interval 120 msec. Record in sensorimotor cortex of rabbit. Arousal by midbrain reticular stimulation.

by several authors.[8,9,11] In brief, there is a depression of surface negative phases of evoked responses and – depending on the stimulus intensity – a more or less marked positivity in their stead. By contrast, after administration of thiosemicarbazide, the depletion of preexisting GABA is signaled by an enhancement of the amplitude of the same negative components; these can be restored to their initial characteristics by intravenous administration of pyridoxine or pyridoxal phosphate.[9]

Before either of these drugs was employed, high frequency reticular stimulation at intensities adequate to induce desynchronization of the electroencephalogram caused no significant changes in the characteristics of local cortical and transcallosal evoked responses. At the same intensities of reticular stimulation and in the same preparations, recruiting and augmenting responses were markedly decreased in amplitude, especially the first ones, often being flattened out completely. Recruiting responses recorded from the nucleus ventralis anterior, the head of caudate nucleus, or the dorsal hippocampus were also found to be very markedly depressed by reticular stimulation, in agreement with earlier findings.[4]

When GABA was applied topically on the cortex, no further significant changes could be induced by reticular stimulation in any of the responses. When depletion of GABA was induced by intravenous or topical administration of thiosemicarbazide, the amplitude of the negative, and often of the initial positive, components of the various evoked potentials was markedly increased. At this time, reduction or abolition of the recruiting and augmenting responses was not altered, that is, responses were depressed to the same level as they had been before the drug (see illustration). Likewise, responses like the local cortical and transcallosal responses, magnified by the effects of the carbazide, were equally unchanged by reticular stimulation as was the case in the initial controls.

DISCUSSION

The possibility that GABA, as a substance presumably involved in some form in inhibitory processes, might be a factor in the mechanisms of electrocortical arousal was first entertained seriously when it was noted that its topical application on the cortex caused changes in the recruiting responses resembling those induced by arousal. However, reduction of other cortical responses by the drug, including those not affected by arousal, suggested that more than a single mechanism was involved. Critical observations, seemingly making it impossible to account for electroencephalographic arousal in terms of involvement of GABA as an inhibitory transmitter, were obtained when no impairment of the blocking effect of arousal upon recruiting responses oc-

curred even after GABA depletion by thiosemicarbazide.[12–14] The low levels of GABA obtained sixty to ninety minutes after the injection of the carbazide[12,13] might be expected to have some functional manifestation, were GABA an essential substance in the arousal process.

The present observations raise some questions as to whether blocking of recruiting responses during electroencephalographic arousal occurs within the cortex or subcortically, within the thalamus. Failure of electroencephalographic arousal to influence other surface-negative cortical potentials may favor a thalamic locus of action. In agreement, recruiting responses recorded from the thalamic nucleus ventralis anterior were reduced during electroencephalographic arousal to a degree equivalent to those recorded in the cortex. Whichever level further study may determine is the locus, the intravenous administration of thiosemicarbazide in many of the present experiments would have served to deplete GABA as an inhibitory substance just as effectively within the thalamus as in the cortex.

SUMMARY

1. A study has been made of the effects of electrocortical arousal upon some evoked potentials in rabbits immobilized with gallamine triethiodide (Flaxedil) and anesthetized locally at painful points. The resting electroencephalogram of these preparations is that of light sleep, from which the animals could be aroused easily by afferent or midbrain reticular stimulation.

2. Recruiting and, in lesser degree, augmenting responses were reduced or disappeared altogether during electroencephalographic arousal; local cortical and transcallosal responses were not affected.

3. The possibility that gamma-aminobutyric acid (GABA) was involved as an inhibitory substance in this reduction seems unlikely since arousal effects showed no impairment when GABA was depleted from the brain by administration of thiosemicarbazide.

The authors wish to acknowledge the technical assistance of Mrs. Ruth Shull, Miss Georgena Sherer, and Mr. Rod Ferreira.

REFERENCES

1. Purpura, D. P.: Observations on the cortical mechanism of EEG activation accompanying behavioral arousal. Science 123:804, 1956.
2. Purpura, D. P.: A neurohumoral mechanism of reticulo-cortical activation. Am. J. Physiol. 186:230, 1956.
3. Evarts, E. V., and Magoun, H. W.: Some characteristics of cortical recruiting responses in unanesthetized cats. Science 125:1147, 1957.
4. Mopuzzi, G., and Magoun, H. W.: Brain stem reticular formation and activation of EEG. Electroencephalog. & Clin. Neurophysiol. 1:455, 1949.
5. Brookhart, J. M., and Zanchetti, A.: The relation between electro-cortical waves and responsiveness of the cortico-spinal system. Electroencephalog. & Clin. Neurophysiol. 8:427, 1956.
6. Bremer, F.: The neurophysiological problem of sleep. In Brain Mechanisms and Consciousness. Oxford, Blackwell Ed. 1954.
7. Grundfest, H.: General problems of drug actions on bioelectric phenomena. Ann. New York Acad. Sc. 66:537, 1957.
8. Purpura, D. P., Girado, M., and Grundfest, H.: Mode of action of aliphatic amino-acids, on cortical synaptic activity. Proc. Soc. Exper. Biol. & Med. 95: 791, 1957.
9. Eidelberg, E., Hyman, W., Killam, E. K., and French, J. D.: Effects of gamma-aminobutyric acid and thiosemicarbazide on neocortical evoked potentials. Read at the fall meeting of the American Physiology Society, 1957.
10. Florey, E., and McLennan, H.: Effects of inhibitory factor (factor I) from brain on cerebral synaptic transmission. J. Physiol. 130:446, 1955.
11. Iwama, K., and Jasper, H. H.: The action of gamma aminobutyric acid upon cortical electrical activity in the cat. J. Physiol. 138:365, 1957.
12. Killam, K. F., and Bain, J. A.: Convulsant hydrazides; in vitro and vivo inhibition of vitamin B_6 enzymes by convulsant hydrazides. J. Pharmacol. & Exper. Therap. 119:255, 1957.
13. Killam, K. F.: Convulsant hydrazides; comparison of electrical changes and enzyme inhibition induced by administration of thiosemicarbazide. J. Pharmacol. & Exper. Therap. 119:263, 1957.
14. Roberts, E., Rothstein, M., and Baxter, C. F.: Some metabolic studies of gamma-aminobutyric acid. Proc. Soc. Exper. Biol. & Med. 92:796, 1958.
15. Sawyer, C. H., Everett, J. W., and Green, J. D.: The rabbit diencephalon in stereotaxic coordinates. J. Comp. Neurol. 101:801, 1954.
16. Rossi, G. F., and Zanchetti, A.: The brain stem reticular formation. Arch. Ital. de Biol. 95:199, 1957.
17. Verzeano, M., Lindsley, D. B., and Magoun, H. W.: Nature of recruiting response. J. Neurophysiol. 16: 183, 1953.

Radioactive phosphate concentration in the cerebral cortex

Louis Bakay, M.D.

PREVIOUS EXPERIMENTS[1] revealed that after administration of radioactive phosphate the highest relative specific activity was obtained in the surface areas of the brain, that is, in the superficial layers of the cortex and in the walls of the ventricles. The pattern of these high concentration areas was the same whether P^{32} was injected in the blood circulation or directly into the cerebrospinal fluid. It was particularly striking that the P^{32} concentration in the cortex declined gradually from the superficial to the deeper laminae. It was felt that an attempt should be made to correlate this observation with our present knowledge of cortical histology and histochemistry.

MATERIALS AND METHODS

Observations were made in cats and in man. In cats, an isotonic solution of P^{32} was injected intravenously in the form of $Na_2HP^{32}O_4$ in the amount of 100 μc. per kilogram of body weight. Larger doses could have caused cellular, axonal, and myelin degeneration, particularly in the cortex.[2] The animals were killed thirty minutes to twenty-one days after injection either by air emboli or cutting of the medulla. Shortly before death, the bone of the calvarium was removed in order to facilitate the immediate removal of the brain. Blood and cisternal fluid samples were taken routinely. The brain was immediately frozen and kept at $-20°$ C. throughout the subsequent laboratory work. Contact radioautographs of 100 to 500 μ cross sections of the brain were prepared on Kodak No-Screen x-ray film. At the same time, various parts of the brain, particularly the various cortical layers of the parietal lobe, were isolated from the frozen specimen and their radioactivity determined by end-window G-M counters. The individual samples were weighed on aluminum planchets and ashed before counting.

A standard place of examination was selected. This was located in the upper midparietal lobe and involved the marginal and suprasylvian gyri medially and laterally from the lateral sulcus. Examination of the native frozen sections showed that the average thickness of the cortex was 1.6 mm. at the top of the gyri and 1.2 mm. at the bottom of the sulcus. Fixation of the brain in 10% formalin for histologic purposes resulted in a reduction of 20% between two measured points, and it was assumed that all cortical layers would be equally affected by this shrinkage. A corresponding correction was carried out after measuring the width of various layers in the fixed specimens. The width of various laminae of the parietal cortex was measured under the microscope as well as in microphotograms with accurately determined magnification in cresyl-violet-stained sections. Layer I averaged 162 μ (10%) of the total cortical thickness; layer II, 130 μ (8%); layer III, 420 μ (26%); layer IV, 129 μ (8%); layer V, 360 μ (22%); and layer VI, 419 μ (26%). These values are similar to those found in rats with the exception of layer III, which is wider in the cat.

The final data were computed in counts per minute per milligram of fresh tissue. The aim in this work was to determine P^{32} concentration in various layers of the cortex, and, therefore, a strict comparison between the various experiments was not necessary. It was found that a rough estimation could be done by computing the data to the P^{32} level in the plasma and correcting this again with the time elapsed from the P^{32} injection, using an average P^{32} plasma time-concentration curve. The total, as

From the Department of Neurosurgery, Massachusetts General Hospital, Boston.

This work was supported by research grant B212-C5 from the National Institute of Neurological Diseases and Blindness, the National Institute of Health, and the U. S. Public Health Service.

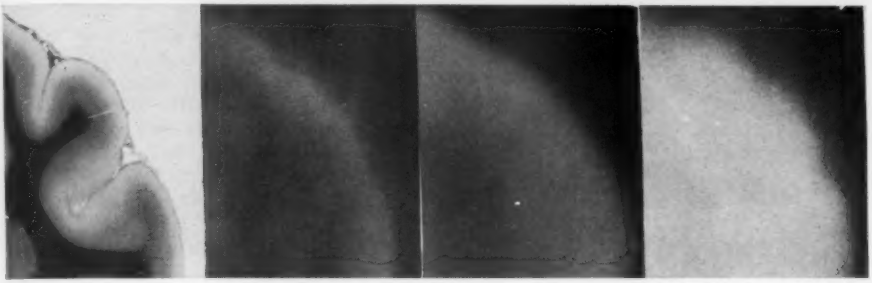


Fig. 1. Radioautographs of cat cortex two hours, three days, and three weeks after intravenous injection of 0.3 mc. of P^{32}. The white areas correspond to darkening on the original radioautographs and indicate high P^{32} concentration. Finer details of the original autographs are lost in the print. Exposure time was ten days. (× 3)

well as the inorganic phosphate content of the combusted cerebral cortex, was determined by the method of Fiske and Subbarow. We were unable to determine the concentration of various phosphate fractions in the individual laminae of the cortex, and, for this reason, it was impossible to give our data in terms of specific activity.

Human data were obtained mostly from patients with brain tumors who died at various intervals after administration of 1 to 2 mc. of P^{32} for tumor localization. The brain was removed after death and processed as described above. The lapse of time between death and removal of the brain at autopsy was of considerable length, varying from two to eight hours. Particular attention was given to the upper mid-portion of the supramarginal gyrus (Brodmann's area 40) for comparative purposes. The width of the cortex in unfixed frozen sections averaged 4.3 mm. at the top of the convolution and 3.4 mm. underneath the midparietal sulcus. The cortical lamination differed from that of the cat because of the phylogenetically increasing development of the supragranular layers (laminae II and III). Lamina I averaged 10% of the cortex; lamina II, 11%; lamina III, 31%; lamina IV, 12%; and laminae V and VI, 36%.

RESULTS

Cats. Radioautographs showed the greatest concentration of P^{32} in laminae I and II of the parietal cortex within the first hour after intravenous injection. Later on, laminae III and IV and, eventually, the entire cortex and the underlying white matter became involved (Fig. 1). However, at no time was the increased concentration limited to layers or to the cortex alone; it gradually progressed into the white matter. Nevertheless, the gradient of concentration from surface to depth remained quite striking throughout these experiments, although it decreased with the elapse of time from injection.

The initial penetration of P^{32} limited itself to the superficial layers of the top of the convolutions and reached corresponding layers at the bottom of the sulci much later, when the zone of penetration had advanced to this level of depth. Consequently, high concentration of the isotope in the supragranular layers at the bottom of a sulcus was attained only when it had already passed the granular or infragranular layer at the top of the gyrus. This phenomenon clearly indicated that factors other than the metabolism of certain individual cortical layers accounted for the exchange of P^{32} between plasma and brain.

Direct determination of the radioactivity also bore out this observation (Fig. 2), even though the isolation of cortical layers was not entirely successful due to the small diameter of the cat cortex. Lower layers of the top of the gyri were somewhat mixed by higher layers from the declining portions of the gyri. The contamination of the specimens with subcortical white matter was not significant, as the superior and midparietal lobes of the cat contain not more than 30% of white matter.

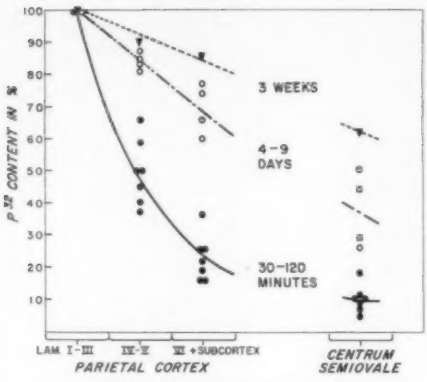


Fig. 2. Relative concentration of intravenously injected P^{32} in various layers of the feline cortex. The isotope content of laminae I to III is arbitrarily designated as 100%.

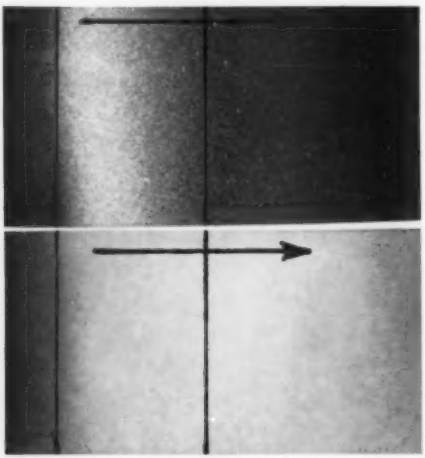


Fig. 3. Radioautographs of a normal *(upper)* and pathologic (toxic encephalitis, *lower)* parietal cortex of man four days after intravenous injection of 2 mc. of P^{32}. Area between lines represents the width of the cortex. Arrows indicate the direction of isotopic diffusion. The lower autograph indicates generally increased permeability of the blood-brain barrier in severe encephalitis. Exposure time was twelve days. (× 4)

Human beings. This material could not possibly include observations on the earliest phase of phosphate exchange between plasma and cortex. However, the later phases were well represented. The relative thickness of the cortex made the isolation of various layers easier and more accurate than in the studies with cats. The findings were essentially identical with those of the animal experiments both by radioautography (Fig. 3) and tissue analysis (Fig. 4). It was interesting that the unavoidable time lag between death and autopsy did not interfere with the topical concentration of P^{32}. This observation was made by comparing the data with those obtained in cats where the brain was immediately removed and frozen. This seems to indicate that the free entrance of P^{32} into numerous metabolic reactions, which comes to an end almost immediately at the time of death, does not affect the validity of observations confined to the topographic distribution of the isotope.

Figure 5 shows that the penetration of P^{32} advanced at the same rate from the surface into the lower parietal cortex and from the corresponding lateral ventricle into the adjacent white and gray matter. It is also shown that the cortex of the insula and the depth of the sulci contained only little more than the deep white matter and very much less than the cortex of the superficial gyri.

DISCUSSION

How can we explain this striking pattern of P^{32} distribution in the cortex? My previous experiments,[4] as well as those of Brierley,[5] rule out the importance of the capillary network in the laminar distribution of P^{32}. There was no direct relationship between the specific activity and capillary population in any given region.

The next step was to investigate the relationship between the rate of phosphate exchange and the volume of cells in any particular layer of the cortex. It could be assumed that there is a direct correlation between phosphate uptake and cellularity because of the importance of this ion in cellular metabolism. This problem is greatly hampered because of our inadequate knowledge of the total volume of cells. There is also the question of the relative metabolic activity of neurons and various

glial cells. It is generally assumed that the total volume of nerve cell bodies in the cortex (although not the total number of cells) equals that of the glial cell bodies exclusive of their processes. There is also growing evidence that the part played by neuroglia in cortical metabolism is by no means negligible. Pope[6] came to the following rough approximation concerning relative volume fractions in the cortex. Neurons, especially their dendrites, make the largest single contribution to the intracellular phase, followed by the protoplasmic astrocytes. Roughly one-third of the cortical volume is due to neurons, one-third to the extracellular fluid compartment, and one-third to the glial cells and blood vessels.

By breaking down the available data in terms of laminar distribution, the cell frequency profiles, as shown by Campbell,[7] reveal no clear-cut relationship with our P^{32} curves. The cell frequency profile, as well as scanning densitometry, showed considerable variation between various cortical laminae. There is a general decline in total nerve cell volume from laminae III to VI. However, the range is relatively slight and does not approach the much wider range of P^{32} distribution. These data do not explain at all the high P^{32} concentration in laminae I and II.

Recent publications contain data on the architectonic distribution of enzymes in various layers of the cortex. Pope's investigations[8] showed that the superficial layers contain a good deal more of some of these enzymes than the layers underneath them. Adenosinetriphosphatase, diphosphopyridine nucleotide, and, particularly, cytochrome oxidase form a gradually decreasing concentration curve from laminae I to VI. Similar, steadily decreasing concentrations of acid phosphatase and fumarase from laminae I to VI were found by Robins and associates[9,10] in the motor cortex of monkeys. In spite of some inconsistencies, the concentration of diphosphopyridine nucleotide and cytochrome oxidase does, to a certain extent, parallel the mass of the nerve cell bodies and their protoplasmic expansions at the different cortical architectonic levels. It is interesting that those enzymes regulating cellular respiration and phosphorylation form a distribution pattern superficially resembling the pattern of phosphate deposition.

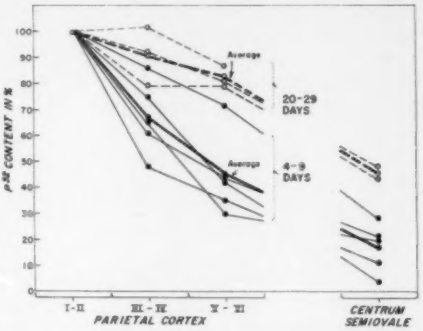


Fig. 4. Relative concentration of intravenously injected P^{32} in various layers of the human cortex. Concentration in the superficial layers is taken to be 100%.

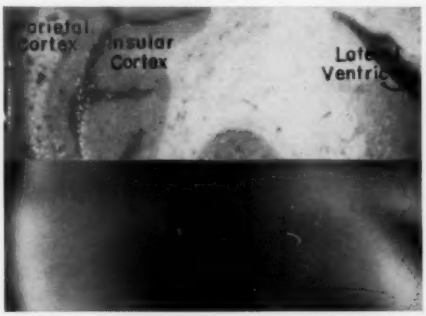


Fig. 5. Section of human brain with corresponding gross radioautograph four days after administration of 2.6 mc. of P^{32}. Exposure time was forty-eight hours. (× 2.2)

It is also interesting to compare the distribution of P^{32} with another primarily intracellular ion, radioactive sulfur (S^{35}). Cohn, Gaitone, and Richter[11] showed that S^{35}-labeled methionine penetrates the brain at its entire depth and with great rapidity when injected into the cerebrospinal fluid. Considerably larger doses of the tracer were required to obtain a similar effect by intravenous administration, despite the fact that this substance diffuses relatively freely through the peripheral capillaries elsewhere in the body.[12] The pattern of deposit inside the brain is essentially the same whether the tracer is injected intra-

venously or intracisternally. The same observation with P^{32} was made by Bakay.[1] However, the cerebral deposition of S^{35}-labeled methionine differs from that of phosphate; S^{35}-labeled methionine concentrates in the individual cells. Consequently, there is a direct relationship between maximum specific activity and cell population, a relationship which cannot be demonstrated for P^{32}. MacLean and associates,[13] as well as Flanigan and associates,[14] studied the fate of radiosulfur in the brains of rats receiving S^{35}- labeled L-methionine under a variety of experimental conditions by radioautography. Under normal conditions, the cerebral radioautograms showed depths of contrast that gave them a striking resemblance to cell-stained sections. The degree of intensity of radioactivity paralleled the intensity of staining seen in Nissl preparations. Similar findings were reported after intraperitoneal administration of S^{35}-labeled sodium sulfate.[15,16]

The P^{32} concentration of various layers of the cortex at different time intervals can best be explained by assuming that the great bulk of the isotope penetrates its surface from the outside and advances into the deeper layers by diffusion. It is known that the physical process of diffusion of most substances, including isotopes, requires considerable time from their point of entrance into the brain to more remote parts. During this period, their spatial availability does not reflect on their final metabolic utilization. Purely physical diffusion is superimposed on biochemical processes, and the relative importance of the two is difficult to assess. It was pointed out that when the time required for diffusion into the depths of the tissue is longer than that required for metabolic exchange, the physical processes of diffusion become of considerable or even predominant importance.[17]

The rate of phosphate entry into the brain is determined to a great extent by the concentration gradient of P^{32} between plasma and brain, as the blood-brain barrier is not impermeable to this ion. Streicher[18] showed that, if the differential is sufficiently large, the rate of penetration of the ion is not limiting with respect to intracellular phosphorylation. However, this phase is in effect for only sixty minutes or less after the administration of a relatively large dose of tracer.[18] This period coincides temporally with the highest level of specific activity in the plasma. There is now evidence that some of the P^{32} reaches the brain through the cerebrospinal fluid, even after intravenous administration of the tracer.[1,19] The true concentration gradient between cerebrospinal fluid and brain after intravenous injection of P^{32} cannot be determined because of an immediate transfer of the relatively slowly accumulating P^{32} of the cerebrospinal fluid to the brain. Davson[19] pointed out that the apparent slowness of penetration of inorganic P^{32} from plasma to cerebrospinal fluid is probably due to losses to the surrounding nervous tissue after the phosphate has entered the cerebrospinal fluid.

The best description of the process of phosphate exchange in the three-compartmental system of blood, cerebrospinal fluid, and brain is given by Davson:[19]

> Labeled inorganic phosphate passes across the blood-cerebrospinal and blood-brain barriers; because, however, of the rapid losses of labeled phosphate to the metabolizing nervous tissue — where it is incorporated into organic complexes — the concentration in the cerebrospinal fluid is continuously greater than that in the extracellular fluid of the nervous tissue; consequently, there is continuous leakage of this material into the nervous tissue, and the concentration is prevented from rising to its characteristic true steady-state level.

SUMMARY

The laminar distribution of P^{32} was studied by radioautography and by direct tissue analysis in the parietal cortex of the cat thirty minutes to twenty-one days after a single intravenous injection of the isotope. Similar studies were undertaken in the parietal cortex of human brains recovered at autopsy four to twenty-nine days after the administration of P^{32}.

During the first hour, P^{32} concentrates mostly in laminae I and II. Later, the zone of high concentration gradually involves laminae III and IV and eventually the entire cortex and subcortical white matter. The concentration gradient from surface to depth diminishes with time but is still noticeable four weeks after injection.

The pattern of P^{32} deposition cannot be explained by the volume of cells in various cortical laminae. In spite of its superficial resemblance to the laminar distribution of a number

of enzymes, the P^{32} concentration in various layers of the cortex is apparently determined to a great extent by its physical diffusion from the cerebrospinal fluid.

REFERENCES

1. Bakay, L.: The Blood-Brain Barrier with Special Regard to the Use of Radioactive Isotopes. Springfield, Charles C Thomas, 1956, pp. 46-60.
2. Di Virgilio, G., and Lavenda, N.: Effects of radiophosphorus on the central nervous system of rodents. Anat. Rec. 127:410, 1957.
3. Pope, A., Hess, H. H., Ware, J. R., and Thomson, R. H.: Intralaminar distribution of cytochrome oxidase and DPN in rat cerebral cortex. J. Neurophysiol. 19: 259, 1956.
4. Bakay, L.: Relationship between cerebral vascularity and P32 uptake. Arch. Neurol. & Psychiat. 78:29, 1957.
5. Brierley, J. B.: Prolonged and distant effects of experimental brain injury on cerebral blood vessels as demonstrated by radioactive indicators. J. Neurol. Neurosurg. & Psychiat. 19:202, 1956.
6. Pope, A.: The relationship of neurochemistry to the microscopic anatomy of the nervous system. In: Biochemistry of the Developing Nervous System. H. Waelsch, editor, New York, Academic Press, Inc., 1955, pp. 341-349.
7. Campbell, B.: Organization of cerebral cortex; introduction and methodology. J. Neuropath. & Exper. Neurol. 13:407, 1954.
8. Pope, A.: Application of quantitative histochemical methods to study of nervous system. J. Neuropath. & Exper. Neurol. 14:39, 1955.
9. Robins, E., Smith, D. E., Eydt, K. M., and McCaman, R. E.: Quantitative histochemistry of the cerebral cortex; architectonic distribution of nine enzymes in the motor and visual cortices. J. Neurochem. 1:68, 1956.
10. Robins, E., Smith, D. E., and Eydt, K. M.: Quantitative histochemistry of the cerebral cortex; analyses at 50 μ intervals compared with analyses by architectonic layers in the motor and visual cortices. J. Neurochem. 1:77, 1956.
11. Cohn, P., Gaitonde, M. K., and Richter, D.: Localization of protein formation in rat brain. J. Physiol. 126:7 P, 1954.
12. Kovach, R. D., Freis, E. D., Schnaper, H. W., and Lilienfield, L. S.: Transcapillary exchange of S^{35}-labeled L-methionine in the human forearm. Proc. Soc. Exper. Biol. & Med. 94:88, 1957.
13. MacLean, P. D., Flanigan, S., Flynn, J. P., Kim, C., and Stevens, J. R.: Hippocampal function: tentative correlations of conditioning, EEG, drug, and radioautographic studies. Yale J. Biol. & Med. 28:380, 1956.
14. Flanigan, S., Gabrieli, E. R., and MacLean, P. D.: Cerebral changes revealed by radioautography with S^{35}-labeled L-methionine. Arch. Neurol. & Psychiat. 77:588, 1957.
15. Boström, H., and Odeblad, E.: Autoradiographic observations on the uptake of S^{35}-labelled sodium sulfate in the nervous system of the adult rat. Acta psychiat. et neurol. scandinav: 28:5, 1953.
16. Ringertz, N. R.: On the sulphate metabolism of the mousebrain. Exp. Cell Res. 10:230, 1956.
17. Garoutte, B., and Aird, R. B.: Diffusion from brain slices in vitro. J. Cell. & Comp. Physiol. 48:167, 1956.
18. Streicher, E.: The entry of $P^{32}O_4$ into the rat brain. J. Cell. & Comp. Physiol. 48:139, 1956.
19. Davson, H.: Physiology of the Ocular and Cerebrospinal Fluids. Boston, Little, Brown & Co., 1956, pp. 249-253.

Electroencephalographic changes in acute brain stem vascular lesions

Walter J. Friedlander, M.D.

IT HAS BEEN WELL ESTABLISHED that destructive lesions located anywhere from the diencephalon rostrally may produce alterations in the electroencephalogram; probably, lesions of the spinal cord do not.[1,2] There has been very little clinical or experimental investigation into the electroencephalographic changes associated with destructive, nontumerous lesions of the intervening brain stem – mesencephalon, pons, and medulla – however, it is apparently assumed that changes do not occur or if they do they are mild and without significance.[3–6] Whether or not there are electroencephalographic alterations, the type and location of any modifications and the particular areas of the skull from which these changes may be recorded should be of both physiologic and clinical interest. This is particularly so because of the recent interest in the entity of basilar and carotid artery insufficiency.

TECHNIC

All cases with possible acute brain stem vascular accidents from two Veterans Administration general medical and surgical hospitals with neurologic services were reviewed. Basis for selection of cases included in this study was:

- Definite clinical findings of an acute vascular lesion of the medulla, pons, and mesencephalon, with some definite objective clinical findings at the time of the first electroencephalogram. All lesions eventually turned out to be thromboses or emboli.
- No evidence at the time of the examination nor any past history to suggest pathology of the diencephalon or more rostral structures.
- No serious systemic disease such as malignant hypertension, severe diabetes, or other endocrine or metabolic disturbances.
- No history suggestive of epilepsy. Episodes of coma that could be associated with the brain stem cerebrovascular accident were admissable.
- No history of serious head injuries or brain surgery.

Arteriosclerosis or even roentgenographic evidence of some calcification of the carotid arteries was not reason of itself for exclusion; disorientation, delirium, or marked memory impairment, however, was reason for exclusion. Both mono- and bipolar electroencephalographic recordings were made.

Although this project was entered into with the idea that a large series would not be difficult to accumulate, by the time the long preliminary list of cases was reviewed, only 31 cases were found that met the criteria for an acute vascular accident involving the brain stem without good reason to suspect other complications that could in turn produce significant electroencephalographic changes. To supplement this list, the literature was searched, but, again, optimism was short-lived; though a large number of various types of brain stem cerebrovascular accidents are reported, either the cases are complicated by other factors or, more often, no electroencephalographic findings are reported.

An attempt was made to localize the pathology longitudinally in the brain stem, that is, the mesencephalon, mesencephalopons, pons, pontomedulla, or medulla. The basis for this localization was purely clinical. Only 2 patients came to autopsy, either because of subsequent extension of the lesion beyond the brain stem (case 17) or because of a new lesion at a later date which was beyond the brain stem (case 1). The danger of making a dogmatic statement about the area of brain involved purely on clinical grounds is well appreciated and should be kept in mind in arriving at any definite conclusions from the

From the Neurology Section, Veterans Administration Hospital, Boston.

material to be presented. It was because of this that no further attempt at anatomic localization was thought feasible. Initially, an attempt was made to localize the lesions also in a transverse plane, that is, superiorly or inferiorly and medially or laterally. This could be done in some cases, but so frequently this could not be done with any good degree of certainty that it was felt unwise to attempt this for it would probably result in increasing the inexactness of our already somewhat tenuous basis for localization of the lesions. Also, when some latitude was given to our imagination in localizing lesions in a transverse plane, no definite correlations were obvious in this series between such localizations and the electroencephalographic pattern.

The various localizations that were attempted were based on the clinical examinations made shortly after the patients were admitted to the hospital and not on the past history. Table 1 lists the patients; their ages; dates of admission to the hospital, and, hence, the approximate date of examination upon which the localization was based; the date of onset of the acute episode; the suspected anatomic localization; and the date and findings of the various electroencephalograms. A summary of the cases is listed in the appendix. A diagnosis of posterior inferior cerebellar artery thrombosis – localized in the medulla – was made when the patient had most of, and no more than, the following involved unilaterally – ambiguus nucleus and descending trigeminal, medial lemniscus, cerebellar, and descending sympathetic fibers.

FINDINGS

Abnormal electroencephalographic findings were found in 9 patients; 2 had borderline electroencephalograms, and the remaining 20 records were interpreted as being within the limits of normal. The most outstanding finding was that electroencephalographic changes, when present, were of a minor or mild degree; in no case was the abnormality considered more than a mild one. The only abnormality found in case 10 was a shift in his alpha rate to 7 cps; findings in case 8 were mildly abnormal because of random and diffuse theta activity. The other abnormal records had some indication of focal or at least unilateral abnormality with or without an increase in the amount of random and diffuse theta. These focal abnormalities were again of the 4 to 7 cps wave variety except cases 25 and 1 where the theta assumed a quasiparoxysmal appearance; no significant amplitude asymmetries were seen. The foci were frontotemporal in one case, occipital in another, posterocentral in a third, and central in a fourth. In the remaining 3 patients, the abnormality could be localized only to one side. The relation between the location of the electroencephalographic abnormality and the anatomic area of pathology is summarized in Table 2. There are too few cases to reach any definite conclusions about the relation between the side of the lesion and the side of the electroencephalographic abnormality, but at least once (case 3), the abnormality was on the side opposite the pathology. In another instance (case 15), it was on the same side as the major involvement of a bilateral lesion. Perhaps it is of some interest that all 4 patients with a definite focus had their lesions in the mesencephalopons or pons.

Opening the eyes blocked the alpha in all 15 cases so tested (Table 1); auditory stimulation in one patient (case 16), in which opening the eyes was not attempted, produced only minor changes. Intermittent light stimulation was used in 7 patients. No paroxysmal electroencephalographic activity was seen, and there was always good and bilaterally symmetrical drive. However, in case 17 and perhaps in case 19, the drive was better delineated anteriorly and temporally than posteriorly; this anterior and posterior difference is just the opposite of what is usually seen with the technic employed in this laboratory. Sleep electroencephalograms were obtained in addition to routine records in 3 patients; all of these were considered normal and contained normal amounts and distribution of sleep spindles and K complexes in response to auditory stimuli (Table 1). A Metrazol-activated electroencephalogram, using a slow, intravenous technic, was obtained in 1 patient (case 15). There were nonfocal, paroxysmal bursts after the administration of 250 mg., which is a response that is observed in only 10% of patients with possible neurologic disease who do not have seizures.[7]

TABLE 1
CASES OF CEREBROVASCULAR ABNORMALITIES IN THIS REPORT

Case	Date of onset	Date of hospitalization	Age	Date of electroencephalogram	Location of lesion	Electroencephalographic findings	Alpha blocked with eyes open
1	1½ years before admission	5-11-51	35	6-14-51	Mesencephaloponto medulla, bilaterally	Abnormal (Table 2)	Yes
				6-21-51		Sleep electroencephalogram, normal	
2	6-19-51	6-19-51	63	6-20-51	Mesencephalon, bilaterally	Normal	
3	2-19-52	2-20-52	54	3-18-52	Pons†	Abnormal (Table 2)	Yes
				3-18-52		Sleep electroencephalogram, normal	
4	3-12-53	3-13-53	61	3-18-53	Medulla	Normal	
5	4-12-53	4-12-53	65	4-16-53	Mesencephalopons	Abnormal (Table 2)	
6	4 months before admission	8-6-53	57	8-11-53	Mesencephalon	Normal	Yes
7	3-12-54	3-19-54	44	3-26-54	Pons	Normal	
8	7-4-54	7-8-54	60	7-9-54	Pontomedulla	Abnormal (Table 2)	
				7-26-54		Abnormal (Table 2)	
9	4-9-55	4-9-55	60	4-15-55	Pontomedulla	Normal, normal response to photic stimulation	Yes
10	4-13-55	5-4-55	59	5-12-55	Pons	Abnormal (Table 2)	Yes
11	5-6-55	5-7-55	61	5-13-55	Pontomedulla, bilaterally	Normal	
12	1-19-56	1-24-56	60	1-25-56	Pons, bilaterally	Borderline because of a few random theta waves	Yes
13	2-24-56	2-24-56	59	3-2-56	Mesencephalopons, bilaterally	Normal	
14	3-19-56	3-19-56	50	3-21-56	Pontomedulla, bilaterally	Normal	
15	11-7-56	12-7-56	53	12-14-56	Pons, bilaterally	Abnormal (Table 2)	
				1-3-57		Abnormal (Table 2)	
				1-8-57		Sleep electroencephalogram, normal	
				1-10-57		Abnormal (Table 2)	
				1-24-57		Metrazol electroencephalogram, nonfocal paroxysmal bursts after 250 mg.	
16	6-20-57	6-22-57	61	6-28-57	Pons, bilaterally	Abnormal (Table 2), poor but not complete loss of response to auditory stimuli	
				7-2-57		Abnormal (Table 2)	
				7-16-57		Abnormal (Table 2)	
17	4 months before admission	12-6-57	42	12-15-57	Mesencephaloponto Medulla, bilaterally	Abnormal (Table 2)	Yes
				3-1-58		Abnormal (Table 2), better photic drive anteriorly than posteriorly	
				5-7-58		Abnormal (Table 2), normal response to photic stimulation	Yes
18	4 months before admission	3-4-58	69	3-7-58	Pontomedulla, bilaterally	Normal	
				3-28-58		Normal, normal response to photic stimulation	Yes
19	3-7-58	3-8-58	63	3-20-58	Pontomedulla	Borderline because of quasi-paroxysmal activity in central posterior areas bilaterally	
				4-10-58		As before, questionably better photic drive anteriorly than posteriorly	Yes

*Case studies for patients 1 to 21 are given in the appendix.
†Lesions are unilateral unless otherwise noted.

TABLE 1 *(Continued)*

Case	*Date of onset*	*Date of hospitalization*	*Age*	*Date of electroencephalogram*	*Location of lesion*	*Electroencephalographic findings*	*Alpha blocked with eyes open*
20	2-10-58	4-5-58	58	5-22-58	Mesencephalon	Normal, normal response to photic stimulation	Yes
21	4-19-58	4-23-58	51	4-24-58	Medial medulla	Normal, normal response to photic stimulation	Yes
22	3-16-51	3-19-51	54	3-22-51	Posterior inferior cerebellar artery thrombosis, medulla	Normal	
23	5-27-51	6-5-51	59	6-5-51	" " "	Normal	
24	1-15-52	1-30-52	61	2-11-52	" " "	Normal	
25	6-18-52	6-24-52	61	6-27-52	" " "	Normal	
26	6-30-53	7-20-53	66	7-28-53	" " "	Normal	Yes
27	11-22-53	11-22-53	51	12-23-53	" " "	Normal	Yes
28	1-2-54	1-5-54	53	1-6-54	" " "	Normal	Yes
29	10-22-54	10-27-54	58	11-2-54	" " "	Abnormal (Table 2)	Yes
30	10-4-55	10-4-55	63	1-16-56	" " "	Normal	
31	2-21-56	2-21-56	73	2-10-56 2-24-56	" " "	Normal Normal	

DISCUSSION

Of the fairly large amount of experimental work done on the electroencephalographic changes occurring with subcortical destructive lesions, only a few workers discuss lesions that are limited to the mesencephalon or areas caudal to this. Kopeloff and associates[8] injected alumina cream into various subcortical structures of the rhesus monkey and reported the absence of electroencephalographic changes when the material was placed in the tegmentum of the pons or the decussation of the brachium conjunctivum. Lindsley and associates[1] produced large lesions in or actual transections at various levels of the cat's brain stem or upper cervical spinal cord. They reported that transection at C1 or low bulbar areas produced no changes, at the pontobulbar junction produced spindle bursts for about one hour, at the junction of the pons and midbrain produced spindle bursts for several hours, and through the anterior portion of the midbrain produced more persistent spindling. Selective destruction of the medial and lateral lemniscus and the spinothalamic tracts on each side of the midbrain produced but little change in the electroencephalogram. However, they found that mesencephalic lesions which destroyed large areas of the tegmentum reduced or abolished the electrocorticographic pattern of activation and again produced recurring spindles. In subsequent work,[9] they reported that in a destructive lesion of "most of the tegmentum of the brain stem in a plane passing ventrally through the front of the pons while sparing more lateral structures," the electrocorticograms obtained in the awake cat were "slightly more synchronous than normal waking records" but otherwise apparently unremarkable. When these animals appeared asleep, the records were characterized by the normal slow wave pattern but lacked spindle bursts and there was more sharp, fast wave activity than was normally present during sleep. However, if a large lesion were placed in the more anterior midbrain tegmentum so that the diencephalon and lateral afferent paths were again spared, the animal appeared asleep and the electroencephalogram was characterized by slow waves with spindle bursts, that is, a normal sleep record. In these cats with midbrain lesions, ordinary stimuli were without effect during the first four postoperative days, and, even after that, though arousal was possible,

TABLE 2

RELATION BETWEEN ANATOMIC AREA OF PATHOLOGY AND THE ELECTROENCEPHALOGRAPHIC FINDINGS IN CASES WITH ABNORMAL RECORDS

Case	*Age*	*Electroencephalographic findings*	*Location of pathology*
1	35	Questionably abnormal because of questionable left quasi-paroxysmal theta focus	Mesencephalo-pontomedulla, bilaterally
3	54	Mildly abnormal because of a slight but definite theta focus in the right occipital area	Pons, left
5	65	Mildly abnormal because of a low voltage theta and delta focus in the left posterior area	Mesencephalo-pons, bilateral
8	60	Mildly abnormal without focus because of random theta	Pontomedulla, right
10	59	Abnormal because alpha is 7 per second, otherwise normal	Pons, left
15	53	Mildly abnormal because of a left frontotemporal theta focus (electroencephalogram improved over ensuing month)	Pons, bilateral
16	61	Mildly abnormal because of random theta and a theta focus on the left (some slight deterioration in the record over the ensuing two and one-half weeks	Pons, bilateral
17	42	Mildly abnormal because of random theta with a questionable left anterior theta focus, questionable improvement over the ensuing two and one-half months	Mesencephalo-pontomedulla, bilaterally
29	58	Probably abnormal because of quasi-paroxysmal theta in anterior half of head and very questionably greater on the left	Medulla, left

it was very poorly maintained. One of our patients (case 16) who had a mildly abnormal electroencephalogram while awake responded poorly but not incompletely to auditory stimulation. Only 3 of our patients had sleep electroencephalograms, and all were normal with the usual distribution and definition of sleep spindles and K complexes with auditory stimuli. None of our records designed to be obtained while the patient was awake contained an undue amount of sleep. At least 2 of our patients (cases 15 and 3) had a lesion involving the pons; both had sleep spindles and neither had an increase in the fast, sharp wave activity that Lindsley and associates[1] found in cats with lesions in this area. Two factors which of course might mitigate against our findings being similar to those of Lindsley are that our electroencephalograms were obtained a long time after the lesion occurred – two months and one month – and, perhaps, more important, the lesions in our cases were probably smaller than those produced experimentally.

Electrolytic lesions made in cats' midbrain by Knott and associates[10] increased slow activity. These records subsequently returned to normal if the lesion was a small one or if it was unilateral; the abnormality persisted if the lesion was large or bilateral. These authors concluded that it was necessary to destroy significant amounts of the reticular substance in order to get electroencephalographic changes. Freedman and Ferriss[11] produced rostral mesencephalic lesions in cats; they did not give the extent of these, but, from their figures, it must be presumed that the destruction was quite extensive. They then gave the animals Metrazol and found that there was a loss of capacity to respond with high voltage hypersynchronism. Only 1 of our patients (case 15) had a Metrazol activation; his lesion was located bilaterally in the pons and the record was obtained two and one-half months after the occurrence of his cerebrovascular accident. His lesion was caudal to those reported by Freedman and Ferriss that prevented Metrazol-induced hypersynchrony. Hence, the paroxysmal bursts that characterized his Metrazol-activated electroencephalogram might not have been unexpected.

Cate[12] approached the problem somewhat differently; he ligated the vertebral arteries of cats at the level of the first cervical branch and produced acutely a diminution of the electroencephalographic amplitude in all tracings, being more marked posteriorly. However, if the electroencephalogram was obtained several days after ligation, it was normal. This same change was noted even if the ligation was performed as low as the level of C7 or 8.

From this rather small amount of experimental work, it would seem that the higher the lesion is located in the brain stem, the more medial or, at least, the more of the reticular

substance involved, and the sooner the record is obtained after the acute lesion, the more likely it is that there will be electroencephalographic changes. This last factor may be an important reason why more electroencephalographic abnormalities are not found in patients. A perusal of Table 1 shows that there is frequently a considerable lapse of time between the date of onset of the lesion and the date of the electroencephalogram. Also, most of the cases reported in the literature do not give the interval between the onset of the acute cerebrovascular accident and the date that the record was obtained.

As with experimental work, there is a paucity of clinical reports of cases with cerebrovascular accidents limited to the brain stem in which electroencephalograms are reported. Most reviews and textbooks dealing with electroencephalograms or cerebral vascular disease do not even make reference to this entity. Table 3 lists the various individually described cases in the literature offering some clinical and pathologic findings of a vascular accident more or less limited to the brain stem and reporting the electroencephalographic findings. Several other clinical studies should be remarked upon. Gastaut[13] stated that "it appears that infiltrating and destructive lesions of the medulla oblongata and the pons do not appreciably modify cerebral electroengenesis whereas those of the mesencephalon and the diencephalon produce bilateral synchronization in fusiform bursts." He has described[14] two types of electroencephalographic anomalies found in his study of 36 cases of vascular syndromes of the brain stem. One is a modification of the background activity which he describes as being in the sense of a diffuse neural hyperexcitability, for example, fast alpha, desynchronized tracings, beta rhythms, functional spikes in the rolandic and occipital areas, augmented response to photic stimulation, and myoclonic response by recruitment. These findings were seen especially in bulbopontine syndromes. The second type of record has slow delta and theta waves in bursts which may be bilateral or unilateral or multiple spikes or slow waves appearing spontaneously with photic stimulation. These anomalies were seen mainly in the pedunculosubthalamic syndrome. The inclusion of subthalamic lesions in his last group probably leads to their being exluded from our present discussion; individual cases, except in one instance, are not cited. Liberson[15] tells of the report of A. Lundewold who, in an electroencephalographic study of patients in whom alternating consciousness was found after vertebral angiography, found delta activity either unilaterally or bilaterally only when the contrast medium was located in the posterior cerebral arteries. When the contrast medium failed to fill the vessels beyond the basilar artery and its collaterals, the impaired consciousness was unassociated with electroencephalographic changes; however, even in the latter series, no alerting responses with sensory stimuli were observed. Strauss and associates,[16] in discussing their index of asymmetry, reported 3 patients with vascular lesions of the brain stem in whom the indexes did not exceed 1.5. This was within the range of normal.

Obviously not all the cases listed in Table 3 meet all the criteria required for the selection of our own patients; those meeting our criteria for selection and having lesions which could be localized in the same manner used in our study are noted in the table with an asterisk. Since the cases reported in the literature were included because of their brain stem pathology, the assumption has been made that there was no other significant brain pathology, except where commented upon, which might cause electroencephalographic changes. This may not be too realistic on our part.

One serious objection that can be raised to placing very much significance on any of the reported electroencephalographic changes based on clinical work is that most of the patients are elderly and therefore mild electroencephalographic abnormalities should be "taken with a grain of salt." This is a point which has been but recently emphasized by O'Leary[17] and, though without any actual formal investigation, has been borne out by our own experience. How many of the abnormal electroencephalographic readings presented here – in our series of patients and in those in the literature – are due to factors entirely independent of brain stem cerebrovascular accidents cannot of course be determined, but there is no reason to assume that this explanation may not hold for at least some of the cases. This criticism, though a very sig-

TABLE 3
CLINICAL AND/OR PATHOLOGIC AND ELECTROENCEPHALOGRAPHIC FINDINGS IN CASES FOUND IN THE LITERATURE

Reference		Clinical and/or pathologic findings	Localization of lesion†	Electroencephalographic findings
22		Young female, aneurysm of the cerebellar artery		Mild dysrhythmia in temporal area unilaterally
18	*	A 52-year-old male with posterior inferior cerebellar artery thrombosis	Medulla‡	2 to 4 cps activity prominent in all leads, particularly over the right brain; impression: generalized abnormal electroencephalogram: electroencephalogram returned to normal in two weeks.
23		A 17-year-old male with episodes of syncope with nystagmus, diplopia, and blurring vision; diagnosis, transient vertebral artery obstruction		February 1950, normal; July 1950, Dr. Gibbs read electroencephalogram as 14 and 16 cps seizure discharges maximally in the right temporal occipital region with normal background indicative of subcortical type of epilepsy.
13	*	Left oculomotor palsy with crossed hemianesthesia and hemiplegia (pedunculopontine syndrome); pathologically found a small softening in the tegmentum of midbrain and pons	Mesencephalopons	Fusiform bursts of theta at 4 cps bilaterally which were synchronous and symmetrical
24		A 41-year-old with hemiparesis questionably due to a pontine lesion		Slight generalized dysrhythmia
25		A 65-year-old male with evidence of basilar artery insufficiency which became worse after an angiogram that demonstrated a basilar artery thrombosis; at autopsy, found basilar artery, middle, posterior and anterior cerebral arteries thrombosed		Before angiogram, moderate to medium generalized dysrhythmia with slow waves of 6 to 7 cps; no paroxysms; no high voltage; no focal changes
26		Cerebellar thrombosis		Abnormal, probably because of the accompanying arteriosclerotic changes
27	*	A 47-year-old male with destruction of inferior colliculi bilaterally and decussation of the brachium conjunctivum with involvement of medial longitudinal fasciculus, trochlear nucleus; large area of softening bilaterally in brain stem's midline; degeneration of pontine pyramidal fibers	Mesencephalopons, bilaterally	Diffuse symmetrical slow activity
28	*	A 37-year-old male with sudden onset of unconsciousness; pathology, a large hemorrhage in the central pons extending to most caudal part of mesencephalon	Mesencephalopons, bilaterally	Six hours after onset, rather regular rhythm of 8 to 9 cps, 100 μv. with some low voltage fast activity, especially bifrontal and occipital; diffuse bursts of 3 to 4 cps and regular diphasic spikes. Bilateral driving of the cerebral rhythm by flicker. Walter[21] reviewed record and thought it was abnormal as a whole because it showed no signs of any response to stimuli.
20		A 63-year-old male who died after vertebral angiography and showed pathologically, normal cerebral cortex, some diffuse subcortical atrophy, normal basal ganglia and hypothalamus; lower portion of red nucleus deprived of cells, substantia nigra lost a few cells; superior colliculus and lateral geniculate were normal; medial geniculate reduced in size; pontine region including reticular substance was severely damaged; could not find cranial nuclei VI, VII, or VIII but III and IV were normal; pyramidal tracts destroyed for the most part as was cerebellar cortex, dentate and root nuclei		Two electroencephalograms before angiogram were normal; after angiogram, a relatively regular curve was obtained; the alpha activity and voltage remained unchanged; sensory stimuli produced no change, but light, though it did not drive, produced large variations. During sleep, spindles were present but record was again unaffected by painful or auditory stimuli.

TABLE 3 *(Continued)*

Reference	Clinical and/or pathologic findings	Localization of lesion†	Electroencephalographic findings
29a *	A 78-year-old male with transient headaches, vertigo, blindness and mild ataxia; cerebellar tremor on left and bilateral extensor plantar reflexes; diagnosis, basilar artery insufficiency	Medulla, bilaterally	Monopolar record shows that there was some bioccipital slowing in the theta range
b *	A 63-year-old female with left conjugate paralysis, left facial weakness incoordination in all 4 extremities, bilateral spasticity and ataxia; diagnosis, basilar artery insufficiency and arteriosclerotic cardiovascular disease with mild congestive failure	Pons, bilaterally	Normal
c	A 70-year-old female with sudden onset of semicoma and right hemiplegia; improved in next 5 days but followed by a second episode with dysarthria, ataxia, cerebellar tremor of the left hand, deviation of tongue to the right, and right spastic hemiplegia; this episode improved, too; diagnosis, basilar artery insufficiency		After recovery the electroencephalogram showed slow alpha activity
30	A 67-year-old female with a diagnosis of basilar and probably posterior cerebral artery insufficiency		Electroencephalogram was normal between episodes of vascular insufficiency.
31 *	A 58-year-old female with left cerebellar signs and Horner's syndrome; right upper motor neuron signs; pain and temperature modalities decreased on the right (not including the face); and posterior column signs on the right	Pontomedulla	Periods of disorganized frequency with both rapid and slow activity, there being few normal frequency cycles. Localization studied failed to show any cortical dysrhythmia. The changes were uniformly distributed over both hemispheres.
32	Vertebral artery compression upon rotation of head developed in 3 patients.		Normal
33 *	A 23-year-old female with cerebellar signs and tremor on the right during volitional acts and posture holding; paralysis of the left superior rectus; paresis of left inferior and medial rectus; blood but not cerebrospinal fluid Wassermann positive; rheumatic heart disease	Mesencephalopons, bilaterally	No abnormalities
19a	Basilar artery insufficiency followed by a right posterior inferior cerebellar artery thrombosis		Made four weeks later showed 6 per second, rectified sharp waves in the left temporal and intertemporal leads. The record later became normal.
b	Signs of right anterior inferior cerebellar artery thrombosis with later evidence suggesting basilar artery thrombosis		Marked changes with slow and sharp waves in the left temporal and intertemporal leads with some general slowing. Serial tracings while on anticoagulants show improvement.
c *	Old and recent pontine infarcts and thrombosis of the left vertebral artery	Pontomedulla, bilaterally, perhaps	Slowing principally in the temporal regions with shifting lateralization
d *	Pontine and mesencephalic infarctions	Mesencephalopons, bilaterally, perhaps	Before signs of infarctions became evident, showed runs of 3 to 5 cps waves with shifting lateralization. After definite signs of brain stem infarction appeared, there was a worsening of the electroencephalogram with more profound slowing.
34	Basilar artery thrombosis without lesions extending beyond the upper brain stem		Normal

*Cases meeting criteria of author for selection and having lesions which could be localized in the manner of the personally seen cases. Case 20 was not marked with an * only because of the evidence of some diffuse subcortical atrophy.

†Location of the lesion is our presumption based on the clinical and/or pathological findings as noted by the author.

‡Lesions are unilateral unless otherwise noted.

nificant one, does not invalidate the whole study. First, there are patients in their fifth decade or younger included; this type of abnormality does not usually occur in patients this young, but, on the other hand, these abnormalities might be a reflection of arteriosclerotic cerebrovascular disease which these patients may have had regardless of their relatively young age. Second, and more important, in several instances, serial records have shown changes from one electroencephalographic record to another which are highly consistent with an acute vascular lesion. Cohn's case,[18] Tucker's cases (a, b, and d),[19] and our case 15 are examples of this as are perhaps Lunderwold's case[20] and our case 17.

Hence, it must be assumed that an acute vascular lesion of the brain stem may present electroencephalographically as a normal pattern, but this type of pathology is also capable of producing some electroencephalographic change. From our own series at least, the changes are usually only mild in degree and rather nonspecific in character and location. There is the suggestion that lesions involving the pons are more likely to produce electroencephalographic alterations than those in the medulla or mesencephalon, although this may only be a reflection of the small number of purely mesencephalic lesions in our series. However, based on experimental work, medial mesencephalopontine lesions, or at least those involving the more rostral reticular substance, would seem to be capable of altering the electroencephalogram.

Our 3 sleep electroencephalograms and only one Metrazol-activated electroencephalogram do not allow for any evaluation of these special types of records. The number of times we have tried intermittent light stimulation is inadequate, but the fact that 1 and perhaps 2 out of 7 patients had an unusual response – better anterior than posterior drive – suggests that at least this should be investigated further. Although all our patients who opened their eyes during the recording demonstrated blocking of their alpha, the case of Loeb as described by Walter[21] (Table 3) suggests that this, too, may be an avenue for investigation.

Hence, our findings, combined with what could be obtained from other case reports, generally agree with what some others have surmised, that is, that electroencephalographic changes are usually mild when they are found at all. More definite conclusions must await larger clinical series that should include at least several hundred well studied cases.

However, we are not willing to agree to the commonly accepted corollary that the mild electroencephalographic findings are without value. It may well be that the electroencephalogram can offer useful information in the diagnosis of brain stem cerebrovascular accidents, for, under certain circumstances, a mild, nonspecific abnormality or lack of any abnormality can be just as valuable as a gross specific abnormality. For example, based on our series and a review of the literature, the presence of a large, well-defined focus in an electroencephalogram, although not diagnostic,[19] is very suggestive that there is at least some pathology in addition to a brain stem cerebrovascular accident. Also, a marked electroencephalographic abnormality in the presence of clinical signs of brain stem pathology should help the clinician in arranging the entities in his differential diagnosis so that an acute vascular accident should be considered less likely than some other possibility such as a neoplasm. On the other hand, a mildly and focally or nonfocally abnormal electroencephalogram is not at all inconsistent with a brain stem cerebrovascular accident.

CONCLUSION

The electroencephalographic findings in 31 cases of acute vascular accident of the brain stem – mesencephalon, pons, and medulla – have been reviewed.

Mild to minor electroencephalographic findings, nonspecific in location and character, are not infrequently found. Such electroencephalographic findings as have been described are not at all inconsistent with an acute brain stem vascular lesion, whereas a markedly abnormal electroencephalogram should be strongly suggestive of at least some pathology in addition to a vascular accident in the brain stem.

APPENDIX

Case 1. A 35-year-old male with the following clinical findings: vertical nystagmus on upward gaze, horizontal nystagmus on right lateral gaze, right-sided cerebellar signs, in-

volvement of the right descending trigeminal tract, a Horner syndrome on the left, and involvement of left cranial nerves IV, VI, and VII. Lesion localized perhaps in the mesencephalopontomedulla, bilaterally.

Case 2. A 63-year-old male with the following clinical findings: involvement of cranial nerve III bilaterally, paralysis of upward and downward gaze and of convergence, and left-sided pyramidal signs. Lesion localized in the mesencephalon, bilaterally.

Case 3. A 54-year-old male appeared lethargic and had the following clinical findings: rotary nystagmus, involvement of left cranial nerves VI and VII, pyramidal signs on the right and a questional hemihypesthesia on the right. Lesion localized in the pons.

Case 4. A 61-year-old male with the following clinical findings: cerebellar signs on the left and involvement of the descending trigeminal and nucleus solitarius on the left. Lesion localized in the medulla.

Case 5. A 65-year-old male complained of vertigo and had the following clinical findings: bilateral involvement of cranial nerves III, IV, and VI, bilateral cerebellar signs, involvement on the left of the nucleus solitarius and descending trigeminal tract, and pyramidal signs on the right. Lesion localized in the mesencephalopons, bilaterally.

Case 6. A 57-year-old male with paralysis of upward and downward gaze and involvement of right cranial nerve III. Lesion localized in the mesencephalon.

Case 7. A 44-year-old male with pyramidal signs on the left and involvement of cranial nerve VI on the right. Lesion localized in the pons.

Case 8. A 60-year-old male with the following clinical findings: vertical nystagmus, right-sided cerebellar signs, and involvement on the right also of the nucleus ambiguus cranial nerves VI and VII. Lesion localized in the pontomedulla.

Case 9. A 60-year-old male complained of vertigo and appeared lethargic; clinical findings were: nystagmus on left lateral gaze, Horner's syndrome on the left, and left-sided cerebellar signs. Lesion localized in the pontomedulla.

Case 10. A 59-year-old male complained of vertigo and was noted to have double vision on lateral gaze, cerebellar signs on the left, and pyramidal signs on the right. Lesion localized in the pons.

Case 11. A 61-year-old male complained of vertigo and appeared lethargic; he had paralysis of upward gaze, nystagmus on right lateral gaze, hoarseness, cerebellar signs on the left, and a left-sided hypesthesia. Lesion localized perhaps in the pontomedulla, bilaterally.

Case 12. A 60-year-old male with the following clinical findings: bilateral cerebellar and pyramidal tract findings and involvement of the descending trigeminal and cranial nerve VII on the left. Lesion was localized in the pons, bilaterally.

Case 13. A 59-year-old male with the following clinical findings: vertical nystagmus, involvement of cranial nerves III and VI bilaterally and of the right descending trigeminal tract and right cranial nerve VII, and left cerebellar signs. Lesion was localized in the mesencephalopons, bilaterally.

Case 14. A 50-year-old male with the following clinical signs: nystagmus on left lateral gaze, Horner's syndrome on the right, involvement on the right of the descending trigeminal, cranial nerve VII, and nucleus ambiguus, and left-sided pyramidal signs. Lesion was localized perhaps in the pontomedulla, bilaterally.

Case 15. A 53-year-old male with bilateral cerebellar signs and evidence of involvement of left cranial nerve VI. Lesion was localized in the pons, bilaterally.

Case 16. A 61-year-old male presented the following clinical findings on admission: bilateral cerebellar signs and involvement of left cranial nerves VI and VII. Four days after admission, he became semiconscious and had bilaterally constricted pupils and bilateral pyramidal signs; he remained the same until one month later when he became totally unresponsive. The lesion up until this time was localized in the pons, bilaterally.

Case 17. A 42-year-old male with the following clinical findings: pyramidal signs and involvement of the nucleus ambiguus bilaterally and involvement of right cranial nerve III and left cranial nerves XI and XII. There was x-ray evidence of calcification of the basilar artery. Lesion was localized in the mesencephalopontomedulla, bilaterally.

Case 18. A 69-year-old male with the following clinical findings: bilateral cerebellar signs, a Horner syndrome, involvement of the descending trigeminal tract, nucleus ambiguus, nucleus solitarius, and cranial nerve VII on one side and, on the opposite side, involvement of the spinothalamic tract. Lesion was localized in the pontomedulla, bilaterally.

Case 19. A 63-year-old male presented with classic findings of a posteroinferior cerebellar artery thrombosis but in addition had an abducens palsy. Lesion localized in the pontomedulla.

Case 20. A 58-year-old male had involvement of right cranial nerve III and left-sided pyramidal signs. Lesion localized in the mesencephalon.

Case 21. A 51-year-old male had paresis of right cranial nerve XII. Lesion localized in the medulla.

REFERENCES

1. Lindsley, D. B., Bowden, J. W., and Magoun, H. W.: Effect upon EEG of acute injury to brain stem activating system. Electroencephalog. & Clin. Neurophysiol. 1:475-486, 1949.
2. Merlis, J. K., and Watson, C. W.: Electroencephalogram after injury to spinal cord in man. Arch. Neurol. & Psychiat. 61:695-698, 1949.
3. Cobb, W.: The EEG in relation to cerebral pathology localization: VI Congrés Neurologique International. Rev. Neurol. 97:225, 1957.
4. Markovich, S. E.: The value of EEG in cerebrovascular disease. Electroencephalog. & Clin. Neurophysiol. 10:202, 1958.
5. Rohmer, F., Gastaut, Y., and Dell, M. B.: L'E.E.G. dans la pathologie vasculaire du cerveau. Rev. Neurol. 87:93-144, 1952.
6. Roseman, E., Schmidt, R. P., and Foltz, E. L.: Serial electroencephalography in vascular lesions of the brain. Neurology 2:311-331, 1952.
7. Friedlander, W. J.: Clinical evaluation of Metrazol activation in electroencephalography. Neurology 4: 264-270, 1954.
8. Kopeloff, N., Whittier, J. R., Pacella, B. L., and Kopeloff, L. M.: Epileptogenic effect of subcortical alumina cream in rhesus monkey. Electroencephalog. & Clin. Neurophysiol. 2:163-168, 1950.
9. Lindsley, D. B., Schreiner, L. H., Knowles, W. B., and Magoun, H. W.: Behavioral and EEG changes following chronic brain stem lesions in the cat. Electroencephalog. & Clin. Neurophysiol. 2:483-498, 1950.
10. Knott, J. R., Ingram, W. R., and Chiles, W. D.: Effects of subcortical lesions on cortical electroencephalogram in cats. Arch. Neurol. & Psychiat. 73:203-215, 1955.
11. Freedman, D. A., and Ferriss, G. S.: Effect of mesencephalic lesions on Metrazol-induced cortical activity. Neurology 6:173-178, 1956.
12. Cate, J.: L'attività elettrica del cervello dopo occlusione di diverse arterie. Arch. Internaz. Stud. Neur. 1:462-470, 1952.
13. Gastaut, H.: The Brain Stem and Cerebral Electrogenesis in Relation to Consciousness in Brain Mechanisms and Consciousness. Adrian, E., et al., Blackwell, Oxford, 1954.
14. Roger, M. J., Roger, A., and Gastaut, M. H.: Corrélations électrocliniques dans 36 cas de syndromes vasculaires du tronc cerébral. Rev. Neurol. 89:444, 1953.
15. Liberson, W. T.: Review of psychiatric progress 1957: Electroencephalography. Am. J. Psychiat. 114: 593-596, 1958.
16. Strauss, H., Liberson, W. T., and Meltzer, T.: Electroencephalographic studies; bilateral differences in alpha activity in cases with and without cerebral pathology. J. Mt. Sinai Hosp. 9:957-962, 1942.
17. O'Leary, J. L.: Electroencephalogram in geriatric practice. Geriatrics 12:413-420, 1957.
18. Cohn, R.: Clinical Electroencephalography. New York City. McGraw-Hill Book Co., 1949.
19. Tucker, J. S.: The EEG in brain-stem vascular disease. Some observations relating EEG findings to various combinations of infarction and vascular insufficiency. Electroencephalog. & Clin. Neurophysiol. 10:202, 1958.
20. Lundervold, A., Hauge, T., and Löken, A. C.: Unusual EEG in unconscious patient with brain-stem atrophy. Electroencephalog. & Clin. Neurophysiol. 8: 665-670, 1956.
21. Walter, W. G. In discussion of reference 13.
22. Cafieses Molina, F.: Aneurisma de la arteria cerebelosa anteroinferior. Rev. neuro-psiquiat. 14:528-532, 1951, abstracted in Ex. Med. (VIII) 6:585, 1953.
23. Ford, F. R.: Syncope, vertigo and disturbances of vision resulting from intermittent obstruction of vertebral arteries due to defect in odontoid process and excessive mobility of second cervical vertebra. Bull. Johns Hopkins Hosp. 91:168-173, 1952.
24. Hauge, T.: Catheter vertebral angiography. Acta. Radiol. Suppl. 109, 1954.
25. Haugsted, H.: Occlusion of the basilar artery: Diagnosis by vertebral angiography during life. Neurology 6:823-828, 1956.
26. Hines, C. G., Tenney, L. H., and Hughes, J.: Electroencephalographic diagnosis of organic brain disease. U. S. Nav. M. Bull. 42:101-107, 1944.
27. Klingon, G. H.: Caloric stimulation in localization of brain-stem lesions in a comatose patient. Arch. Neurol. & Psychiat. 68:233-235, 1952.
28. Loeb, C., and Poggio, G.: Eletroencephalograms in a case with ponto-mesencephalic haemorrhage. Electroencephalog. & Clin. Neurophysiol. 5:295-296, 1953.
29. Meyer, J. S., Leiderman, H., and Denny-Brown, D.: Electroencephalographic study of insufficiency of basilar and carotid arteries in man. Neurology 6:455-477, 1956.
30. Millikan, C. H., and Siekert, R. G.: Studies in cerebrovascular disease: I. Syndrome of intermittent insufficiency of basilar arterial system. Proc. Staff Meet. Mayo Clin. 30:61-68, 1955.
31. Stead, E. A., Jr., Ebert, R. V., Romano, J., and Warren, J. V.: Central autonomic paralysis. Arch. Neurol. & Psychiat. 48:92-107, 1942.
32. Tatlow, W. F. T., and Bammer, H. G.: Syndrome of vertebral artery compression. Neurology 7:331-340, 1957.
33. Tolosa, A., and Canelas, H. M.: Sindromes do nucleo rubro. A proposito de tres casos com etiologia sifilitica, um dos quais associado a mioclonias velofaringolaringlas. Arq. Neuro-psiquiat. 8:211-226, 1950.
34. Watson, C. W., and Adams, R. D.: The electroencephalogram in its relation to consciousness and responsiveness in destructive lesions of the brain stem: a clinical pathological EEG study of brain stem diseases particularly basilar artery occlusion. Electroencephalog. & Clin. Neurophysiol. 3:371, 1951.
35. Cohn, R., Raines, G. N., Mulder, D. W., and Neumann, M. A.: Cerebral vascular lesions; electroencephalographic and neuropathologic correlations. Arch. Neurol. & Psychiat. 60:165-181, 1948.

Respiratory effects of cerebellar and brain stem stimulation in monkeys

A possible bioassay preparation

Harold D. Cross, M.D., O. Weems Hollowell, M.D., and Jose M. R. Delgado, M.D.

CENTRAL RESPIRATORY PHYSIOLOGY has been studied in anesthetized preparations for many years, but few experiments have been done in the conscious animal. In 1941, Beaton and Magoun[3] localized inspiratory and expiratory centers in the monkey to the region overlying the inferior olivary nuclei; these centers were subsequently found to be less discrete.[14] The control of respiration, especially its automatism, was the subject of much investigation by Gesell[11] and, more recently, by Hoff and Breckenridge.[12,13] Both studies presented evidence that automatism resides within brain stem structures caudal to the pons and is not dependent upon the vagus or pneumotaxic center. Liljestrand's monograph[14] presented a good historic survey of respiratory studies up to 1953.

Magoun and associates[15] stimulated deep cerebellar areas and reported no respiratory findings. Moruzzi,[16] after observing that faradic stimulation of the vermis portion of the anterior lobe of the decerebrate cat inhibited respiratory and vasomotor reflexes, concluded that the cerebellum had some action in circulation and respiration.

Most previous studies have been done in acute preparations. In this paper, we report the respiratory effects evoked by electrical stimulation of the brain stem and cerebellum with electrodes permanently implanted in the conscious monkey. Experiments comparing quality and thresholds of responses were also done with the same animals under anesthesia; validity of previous work for the conscious animal was tested in this way. Since experiments can be repeated in the same animal for months, we believe that these preparations could also be used as a bioassay to test analeptic drugs.

METHODS

The method to construct and implant needles with electrodes has been previously described.[8,10] Employed were 6 immature Macaca mulatta monkeys. In each animal, 2 needles were inserted into the brain stem and/or cerebellum by means of a modified Horsley-Clarke stereotaxic instrument; the needles remained in position up to three months. Each shaft had 6 leads with contact points 2 mm. apart; the surface of each point was about 1,000 sq. μ, and the greatest diameter of the needle was slightly less than 1 mm.

The Horsley-Clarke coordinates selected in the brain stem on the basis of two acute experiments were 2 mm. lateral to the midline, 5 mm. posterior to the interaural line, and 38 to 50 mm. beneath the cortex. The solitary tract is about 40 mm. below the cortex with these coordinates (Fig. 1). Coordinates used for the cerebellum were 10 mm. posterior to the interaural line, 30 to 40 mm. below the cortex, and 2 mm. from the midline. However, histologic studies revealed that 2 of the 3 needles implanted in the cerebellum strayed considerably so that these latter values are probably accurate only for the fastigial and globose nuclei and not for the dentate and emboliform nuclei, which lie more posteriorly.

From the Department of Physiology, Yale University School of Medicine, New Haven, Conn.

This research was supported by the James Hudson Brown Memorial Fund, the National Foundation for Infantile Paralysis, the Foundations' Fund for Research in Psychiatry, and the Office of Naval Research.

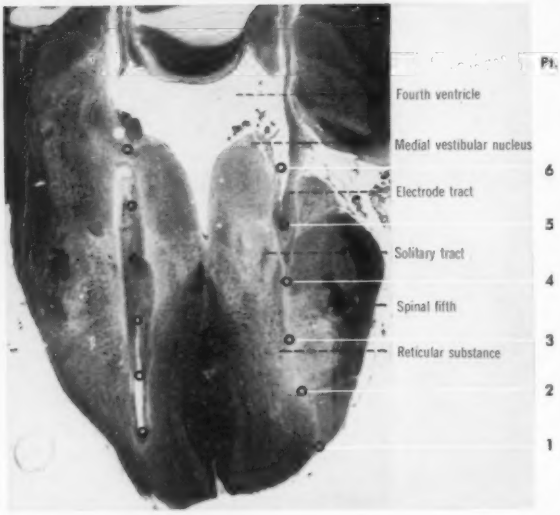


Fig. 1. Coronal section of monkey 3, 5 mm. posterior to the interaural line — resulting in an oblique section due to the angle of the brain stem (Klüver stain, × 8). The small circles represent separate leads. Lowest point is 1. Note the slight reaction along the right tract below the solitary tract and the moderately extensive reaction about both right and left tracts elsewhere. Electrodes had been present ninety-seven days.

Respirations were recorded by having the animal breathe into a closed system connected to a tambour registering respirations on a kymograph. A square wave stimulator was used; stimulation was monopolar and unidirectional. Voltage and amperage were simultaneously monitored with an oscilloscope; since the pulse shape was highly differentiated, we chose to measure the value at its longest duration. Pulse duration was kept constant at 0.25 msec. and the frequency, unless otherwise indicated, was 100 cycles per second. Administration of 40 mg. per kilogram of intraperitoneal pentobarbital maintained surgical anesthesia for about two hours during anesthesia experiments.

LOCALIZATION AND HISTOPATHOLOGY

Localizations of the electrodes and the needle tracts are illustrated in Figs. 1 and 2. Of the 9 needles implanted in the brain stem, 6 were in similar areas in or near the solitary tract. The reaction about the electrodes varied from 0.3 to 1 mm. radial to the lumen, with an average of about 0.6 mm.; the glial scar was composed of a central zone of cellular and collagenous elements. Surrounding the glial scar was a zone of demyelinization and edema in which increased numbers of astrocytes and microglia were seen; neurons of normal appearance were usually adjacent to the glial scar. A sterile abscess was present above the area of stimulation in one animal. A description of the pathologic reaction occurring with chronically implanted pieces of insulated wire has recently been published by Collias and Manuelidis;[6] in our monkeys, however, the reaction in the brain stem was more acute and extensive than that described in their report. This reaction was attributed to the considerable amount of movement which the brain stem supposedly undergoes during the active life of the monkey. Electrical stimulation did not seem to be a significant factor in the amount of reaction, which often was more extensive in nonstimulated than in stimulated areas.[7]

RESULTS

Thresholds of respiratory effects are summarized in the table; responses were classified either as expiratory (E) or inspiratory (I). Expiratory effects were usually characterized by a decrease in inspiratory amplitude and were seen most commonly and consistently in the region of the solitary tract and in lateral reticular areas. Inspiratory response was usually inspiratory apneusis or simply an increase

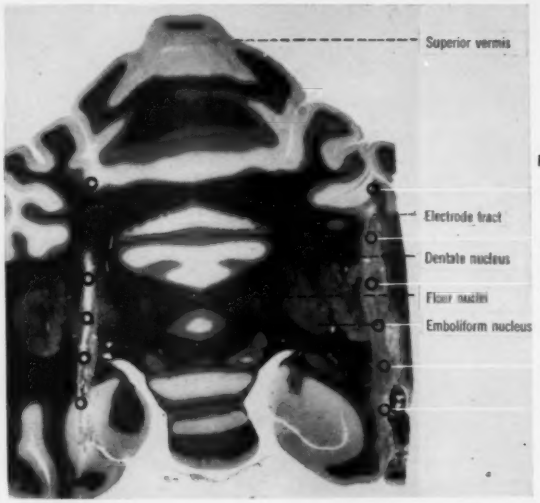


Fig. 2. Coronal section, 10 mm. posterior to the interaural line (Klüver stain, × 6.5). Tracts extend through the dentate and emboliform nuclei on the right and left, respectively. Grossly visible are edema and demyelinization about the tract and hemorrhage at the lower border of the right tract (black area to right of point R1). This hemorrhage is believed to have occurred about twenty-four hours before sacrifice and was probably due to traumatic removal of the electrode shaft.

in amplitude; response was seen chiefly in the cerebellar and medial reticular areas.

CEREBELLUM

In animal 1, needles were inserted through the dentate and emboliform nuclei (Fig. 2). In these structures were 3 contacts of each shaft; the responses obtained from each central point are listed in the table. The most frequent response was an increase in inspiratory amplitude which became apneustic with suprathreshold stimuli. However, this increase would vary during the same experiment, and a severe decrease in amplitude of both inspiration and expiration would occur (Figs. 3*A*, *B*, and *C*). The effects elicited by stimulation of the emboliform nucleus were equally severe (Fig. 3*D*), and, in one experiment, the same sort of variation occurred. In other words, elicitation of respiratory effect was consistent, but the type of response varied.

Escape appeared three to four seconds after onset of stimulation, and the previous respiratory pattern gradually returned. From points in the roof nuclei of animal 2, a consistent and significant increase in inspiratory amplitude occurred without appreciable differences in threshold after stimulation of the animal in both conscious and anesthetized states (see table and Figs. 3*E* and *F*).

BRAIN STEM

Solitary tract. Expiratory-type effects were commonly evoked from this area; however, at some points, the effect was mixed with a decrease in expiratory amplitude, and, at others, with an increase in inspiratory amplitude (Figs. 3*G* and *H*). No significant difference was found in thresholds of animals in conscious or anesthetized states in the first 5 points listed under this area in the table; the threshold in terms of milliamperes (mA) was equal, or within 0.01 mA, at 3 points – higher in the conscious state at one point and higher in the anesthetized state at the other point. An increase in the threshold at some points was noted for seven to ten days, but, at other points, the increase remained essentially constant for three to four weeks.

In animals 3 and 4, gagging, vomiting, and coughing were elicited by stimulation of points in this area during consciousness. The stimulus values were slightly above those used in the respiratory studies. The solitary tract is the same area designated as the vomiting center in the cat by Borison and Wang.[4,19,20] The evoked effect appeared as soon as stimulation was applied and the ejection of food in previously fed animals was considerable. The effect ceased immediately when stimulation

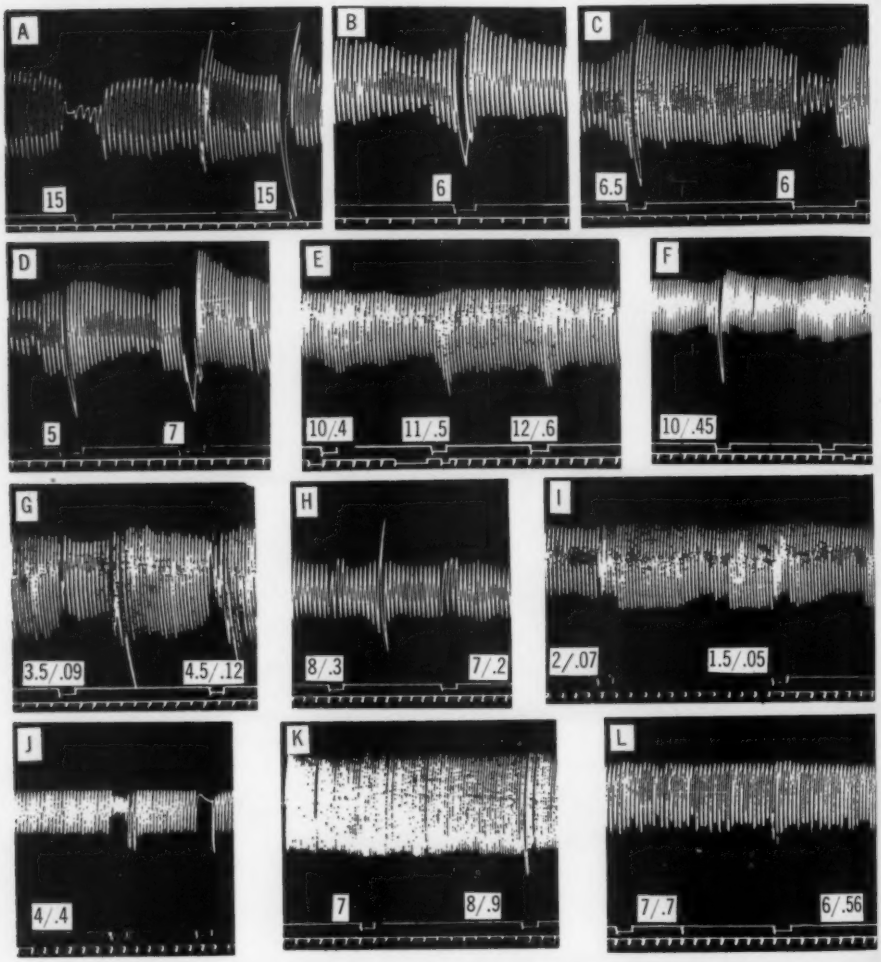


Fig. 3. Respirograms. Inspiration is a down stroke; second line on graph indicates electrical stimulation and third line indicates five second intervals. (*A, B, C*) Animal 1, R5 (dentate nucleus). Awake studies on three different days. Note variation in response from typical inspiratory apneusis to marked inhibition of respiration. (*D*) Animal 1, L5 (emboliform nucleus). Awake study. Severe inspiratory effects were elicited. (*E, F*) Animal 2, L6 (room nuclei of cerebellum). Conscious and anesthetized studies, respectively. Increased inspiratory amplitude was threshold effect. (*G, H*) Animal 3, R6 (solitary tract). Conscious and anesthetized. Decreased inspiratory amplitude was typical effect; however, decrease in rate was noted in anesthetized recording (*H*). (*I, J*) Animal 4, R5 (lateral reticular substance). Conscious and anesthetized. Decreased inspiratory amplitude was usual response in both conscious and anesthetized animals. Anesthetized example shows severe decrease due to suprathreshold stimulation. (*K, L*) Animal 5, L3 (medial reticular substance). Conscious and anesthetized. Usual effect was an increase in inspiratory amplitude.

was stopped, and the monkeys were at once interested in eating ejected substances as well as fresh food. In collaboration with Dr. L. Mihailovic, the action of chlorpromazine was studied in one of the animals. Injection of 1 mg. of chlorpromazine intraperitoneally clearly increased the threshold of the area, and intensities which previously had evoked vomiting (1 mA) produced only gagging when reapplied one hour after injection. This effect was recorded in color motion pictures.

Lateral reticular substance. Expiratory-type effects were generally obtained from points in this area. The quality of the effect in the conscious or anesthetized animal was usually the same, aside from the usual change in the pattern of respiration associated with general anesthesia (Figs. 3*I* and *J*). Comparison of the threshold obtained in conscious and anesthetized states – the first 7 points under this area in the table – fails to reveal any significant difference, with the possible exception of the next to last 2 points in the series (monkey 4, R5 and L3).

The threshold of R3 in monkey 3 was the lowest in this study – 0.015 mA on several occasions. From a few points, no response was evoked with a frequency of 100 cycles per second whereas with 200 to 250 cycles per second, significant effect occurred.

Medial reticular substance. Three needles (9 points) strayed into this area; the effects were usually inspiratory. The quality and threshold of the responses under anesthesia were similar (see table and Figs. 3*K* and *L*). Occasionally, conscious animals uttered crying sounds during stimulation, especially of the lateral reticular areas and the most ventral portion of the brain stem amid pyramidal fibers and the long ascending tracts. Respiratory effects were registered on the kymograph from such movements. Some were apparently entirely motor movement artifact; often, however, the same type of effect, without the sounds, was obtained under anesthesia.

DISCUSSION

Respiratory effects were evoked repeatedly from several areas in the cerebellum and brain stem in conscious and anesthetized monkeys. The effects elicited from cerebellar structures, although studied in only 2 animals, were clear cut, repeated several times, and could be evoked in one anesthetized animal. The variation in response seen in the dentate nucleus from one period of stimulation to another was most curious. The role of the cerebellum in respiration is unknown. One might speculate that the cerebellum acts as a synchronizer of respiratory movements and that stimulation in different phases of the respiratory act would have different over-all effects upon the patterns of response.

The efferent pathways of these responses are also in doubt. Possibilities are the fastigiobulbar tract (uncinate bundle of Russell which originates in the roof nuclei of the cerebellum and terminates in the lateral, vestibular nucleus and in the brain stem reticular substance) and fibers in the brachium conjunctivum and in the brachium pontis.

Snider and associates[18] showed that the facilitation and suppression of movement which may be induced by cerebellar stimulation are mediated by connections with the reticular formation. Bach[1] has pointed out the possibility that bulbar facilitatory and inhibitory systems apply equally well to vasomotor, respiratory, and somatic reflexes. Breckenridge and Hoff[5] proposed that the bulbar systems controlling respiration were identical with the somatic facilitatory and inhibitory reticular system. Bach,[2] however, concluded that the bulbar systems were not identical, but intimately related. Thus, one is led to suspect that cerebellar stimulation influences respiration by way of the facilitatory and suppressor areas in the bulbar reticular formation. These areas are functionally linked – although at present anatomic linkage is poorly understood – to the cerebellum.

The main areas from which respiratory effects were consistently elicited by brain stem stimulation were the solitary tract and the lateral reticular and medial reticular areas. The predominant effect was the same in the solitary tract and lateral reticular areas – a decrease in inspiratory amplitude. This effect has been observed frequently in acute experiments with electrical stimulation.[14,17] In our studies, the effect was the same general type – inspiratory or expiratory – at most points, regardless of the state of consciousness. Likewise, the threshold did not differ significantly in either con-

SUMMARY OF THRESHOLD RESPIRATORY EFFECTS

Area Stimulated	*Animal No.*	*Point No.*	*Type of Response*		*Threshold in mA*	
			Awake	*Anesthetized*	*Awake*	*Anesthetized*
Dentate nuclei	1	R5	I		.25	
Emboliform nuclei	1	L5	I		.13	
Roof nuclei	2	L5	I	I	.15	.18
		L6	I	I	.40	.35
Solitary tract	2	R1	E	E	.15	.15
	3	R6	E	< rate	.09	.20
		L6	E, I	E	.16	.11
	4	R6	E, I	I	.08	.08
		L6	I	I	.07	.06
	3	R5	E		.13	
		L5	E		.15	
	4	R7	E		.11	
		L7	E		.15	
	5	R5	E, I		.08	
	6	R5	E. I		.09	
					—	—
				Mean	.11	.12
				S.D.	.035	.05
Lateral reticular substance	3	R3	E	E	.02	.03
		R5	E	E	.13	.15
		L3	E	I	.09	.08
	4	R3	irregular	0	.20	0
		R5	E	E	.05	.18
		L3	E	E	.07	.11
		L5	E	E	.08	.07
	3	R2	E		.09	
		L2	E		.17	
	4	L2	E		.07	
	6	R2	I		.30	
		R3	I		.12	
					—	—
				Mean	.12	.10
				S.D.	.07	.05
Medial reticular substance	5	L3	I	I	.90	.70
		L5	I	I	.40	.35
		L6	I	incomplete	.20	0
		R3	I		.30	
		R5	E		.13	
		R6	I		.25	
	6	L3	0			
		L5	I		.15	
		L6	0			
					—	—
				Mean	(too few; too great	
				S.D.	variability)	

I: inspiratory type of effect
E: expiratory type of effect
O: no effect

R: right side
L: left side

scious or anesthetized animals. The effect, more clear cut under anesthesia and often modified, usually remained the same basic type. Occasional variations in response to stimulation of apparently identical areas in different animals have also been found by those using only acute preparations.[14,15]

Although crying sounds occurred with stimulation of some points in the lateral reticular areas in conscious animals, respiratory effects were obtained from these same points under general anesthesia. Apparently, although pain fiber stimulation plays a role in the augmentation of respiratory effects, the effect per se can be elicited without these sounds under anesthesia.[9]

The apparent failure to find significant differences in thresholds of respiratory responses obtained in conscious and anesthetized animals was unexpected. Whether this failure could be attributed to the level of anesthesia and/or the specific anesthetic agent used, we cannot say. If different agents do cause detectable differences in thresholds, then this technic may be of value in determining more specifically the site and mechanism of action of some drugs which act on different levels of the central nervous system. For the purpose of evaluating the possible effect of drugs on the threshold of a given effect, the tissue reaction about the electrode would be of little significance if the threshold were determined in the conscious animal just prior to each drug study. In view of the extent of this reaction with chronic implantation, an electrode with a larger surface area – 1 mm. scraped bare wire – would be equally satisfactory for localization and would tend to minimize polarization problems.

SUMMARY

In conscious monkeys with permanently implanted electrodes, respiratory effects were evoked by electrical stimulation of solitary tract and medial reticular substance. Stimulation of dentate and emboliform nuclei of the cerebellum also evoked respiratory responses.

Vomiting was repeatedly induced by electrical stimulation of the solitary tract.

Respiratory effects and thresholds were generally reliable, and it is suggested that this method can be used for bioassay of drugs which have respiratory effects.

In general, use of anesthesia did not modify thresholds or evoked responses. Results obtained in acute experiments, therefore, may be applied to unanesthetized animals.

The help of Dr. L. Mihailovic during these experiments and the technical assistance of Miss Beatrix Egli are warmly acknowledged.

REFERENCES

1. Bach, L. M. N.: The role of the bulbar facilitatory and inhibitory systems in vasomotor and respiratory activity. Fed. Proc. 7:4, 1948.
2. Bach, L. M. N.: Relationships between bulbar respiratory, vasomotor and somatic facilitatory and inhibitory areas. Am. J. Physiol. 171:417, 1952.
3. Beaton, L. E., and Magoun, H. W.: Localization of the medullary respiratory centers in the monkey. Am. J. Physiol. 134:177, 1941.
4. Borison, H. L., and Wang, S. C.: Functional localization of central coordinating mechanisms for emesis in the cat. J. Neurophysiol. 12:305, 1949.
5. Breckenridge, C. G., and Hoff, H. E.: Pontine and medullary regulation of respiration in the cat. Am. J. Physiol. 160:385, 1950.
6. Collias, J. C., and Manuelidis, E. E.: Histopathological changes produced by implanted electrodes in cat brains. J. Neurosurg. 14:302, 1957.
7. Cross, H. D.: Respiratory effects evoked in the awake monkey by brainstem and cerebellar stimulation. Thesis, Yale Univ. School of Medicine, 1957. Chapter on Localization and Histopathology.
8. Delgado, J. M. R.: Permanent implantation of multilead electrodes in the brain. Yale J. Biol. & Med. 24:351, 1952.
9. Delgado, J. M. R.: Cerebral structures involved in transmission and elaboration of noxious stimulation. J. Neurophysiol. 18:261, 1955.
10. Delgado, J. M. R.: Evaluation of permanent implantation of electrodes within the brain. Electroencephalog. & Clin. Neurophysiol. 7:637, 1955.
11. Gesell, R., Bricker, J., and Magee, C.: Structural and functional organization of the central mechanism controlling breathing. Am. J. Physiol. 117:423, 1936.
12. Hoff, H. E., and Breckenridge, C. G.: Levels of integration of respiratory patterns. J. Neurophysiol. 15:47, 1952.
13. Hoff, H. E., and Breckenridge, C. G.: Intrinsic mechanisms in periodic breathing. Arch. Neurol. & Psychiat. 72:11, 1954.
14. Liljestrand, Åke.: Respiratory reactions elicited from medulla oblongata of the cat. Acta physiol. scandinav. (suppl. 106) 29:321, 1953.
15. Magoun, H. W., Hare, W. K., and Ranson, S. W.: Electrical stimulation of the interior of the cerebellum in the monkey. Am. J. Physiol. 112:329, 1935.
16. Moruzzi, G.: Paleocerebellar inhibition of vasomotor and respiratory carotid sinus reflexes. J. Neurophysiol. 3:20, 1940.
17. Pitts, R. F., Magoun, H. W., and Ranson, S. W.: Localization of the medullary respiratory centers in the cat. Am. J. Physiol. 126:673, 1939.
18. Snider, R. S., McCulloch, W. S., and Magoun, H. W.: A cerebello-bulbo-reticular pathway for suppression. J. Neurophysiol. 12:325, 1949.
19. Wang, S. C., and Borison, H. L.: The vomiting center. Arch. Neurol. & Psychiat. 63:928, 1950.
20. Wang, S. C., and Borison, H. L.: The vomiting center: its destruction by radon implantation in dog medulla oblongata. Am. J. Physiol. 166:712, 1951.

Number of axons and myelin sheaths in adult human medullary pyramids

Study with silver impregnation and iron hematoxylin staining methods

William DeMyer, M.D.

THIS STUDY ESTIMATES the number of axons and myelin sheaths in the medullary pyramids of adult human beings with structurally normal brains. Classically, myelin sheaths have been recognized as iron hematoxylin or osmium tetroxide staining cylinders visible with the light microscope. Reviewing the concept of myelination, Schmitt and Geschwind[1] and Fernandez-Moran[2] conclude that, depending on definitions and methods of study, all nerve fibers might be considered as having myelin coverings. In this paper, the term *myelin sheath* refers to the hematoxyphilic sheath identifiable by ordinary light microscopy.

MATERIAL AND METHODS

Medullas were collected at Indiana Medical Center from all autopsied patients fulfilling the following criteria: [1] The patient's age was between 18 and 69 years at the time of death. [2] Death occurred from some acute process. [3] The history indicated that the patient was nonpsychotic and nonalcoholic and had suffered no structural disease of the central nervous system. [4] On physical examination, the patient had no evidence of central nervous system disease.

Patients whose blood pressure had been greater than 160/100 mm. Hg or with severe arteriosclerosis and most patients with chronic debilitating illnesses such as pulmonary insufficiency, congestive heart failure, or cancer were excluded. Preliminary study showed that chronically ill patients often have degenerating nerve fibers even without formal signs of neurologic disease. A few chronically ill but adequately nourished patients who died suddenly of some acute complications were included, however. As determined by gross and microscopic examination, all brains were free from congenital or acquired abnormalities. Of about 2,000 autopsied patients, 25 met the above criteria; 4 of these had to be excluded – 2 because their tissues failed to stain properly and 2 because large circumolivary bundles disturbed the longitudinal orientation of the pyramidal fibers, precluding accurate census.

After the brains had fixed four weeks in 4% formaldehyde, frozen sections of the medullas were made at the junction of the middle and lower thirds of the inferior olivary nucleus. Spielmeyer's method was used to stain myelin sheaths, except that the hematoxylin was freshly prepared and, after preliminary destaining of the sections in iron alum, destaining was completed in Lillie's borax ferricyanide.[3] For axon impregnation, Hortega's silver carbonate[4] was used. The Luxol fast blue stain was done according to the directions of Margolis and Pickett.[5]

Complete details of the fiber counting method are given elsewhere.[6] Using 20-X oculars with an ocular grid and an oil immersion objective, sufficient random samples were counted until the estimate of the true mean number of fibers per sampling unit was expected to be within ± 5% of the true value on 95% of the specimens as shown by the following confidence interval:

$$P\left(\bar{X} + t_{1/2\alpha}\frac{S_X}{\sqrt{N}} < \mu < \bar{X} + t_{1-1/2\alpha}\frac{S_X}{\sqrt{N}}\right) = 1-\alpha$$

The symbol α stands for an arbitrarily chosen

From the Department of Neurology, Indiana University School of Medicine, Indianapolis.

confidence level; throughout this paper, α is 0.05. To achieve this accuracy, about 8,000 fibers or about 175 samples $400\mu^2$ had to be counted. The axons and myelin sheaths were counted in separate sections, without prior knowledge of the census results for the companion section. The counts were all done on the left pyramid.

ANATOMICAL CONSIDERATIONS

The medullary pyramid is distinguished from adjacent structures and tracts by the unique spectrum of its thick and thin fibers. It has a fairly sharp boundary superolaterally against the uniformly thin periolivary fibers. Superiorly, the medial lemniscus is distinguished by its fairly uniform spectrum of medium-sized fibers. Although the corticospinal fibers of the pyramid are fairly well confined to the pyramidal border, as outlined by inspection of the fiber spectrum, a number of cortical efferent bundles sometimes infiltrate the medial lemniscus; this is shown by the degeneration of these bundles in patients dying with cerebral lesions. Small and distinctly separate lemniscal bundles were disregarded; those large and closely packed at the pyramidal border were included in the counts.

Slicing through the pyramids are arcuate bundles, easily excluded from the census by their uniform diameters and oblique or transverse course. Occasionally, isolated transverse fibers appear, but, since they seem to be extraneous or collateral, they are excluded. Remaining are the fibers taking a more or less longitudinal course, running in company with other fibers through the thickness of the section. These fibers constitute the object of the census. If the human medullary pyramids contain ascending fibers, as has been claimed by Nathan and Smith,[7] they cannot be distinguished by morphological examination and must be included.

RESULTS

Table 1 summarizes the raw data and Table 2 the statistical analysis. The mean number of axons in the left medullary pyramid is 1,087,200 for 21 specimens; the range is from 749,900 to 1,391,100. The mean number of myelin sheaths is 1,033,400 for 12 specimens. The mean ratio of myelin sheaths to axons is 93.8%, with a range of 80.4 to 104.1%. The excess of myelin sheaths over axons in specimens 5 and 11 can be explained by the variability of the totals owing to random sampling.

By use of Student's t test, the following subgroups were compared: axon and myelin sheath totals for the 5 oldest versus the 5 youngest specimens and for the 6 female and 15 male patients; mean number of axons only for the 3 Negroes versus the 18 whites; and the percentage of myelinated fibers in the 5 specimens with the highest axon totals versus the 5 with the lowest axon totals in those specimens where both censuses were done. None of these subgroups differ significantly at the 0.05 level from the whole group or from the subgroups they were compared with; the groups were small, however, and the differences would have had to be large to achieve significance.

To test the reliability of the census method, the number of axons in 1 specimen and the number of myelin sheaths in 3 others were reestimated, giving a check on 4 different counts. The second estimates were made on different sections without prior knowledge of the initial results, with one to two years intervening between the first and second counts. Since the maximum difference on recounting was 6% and the mean difference 3%, the results of the second count are well within the expected limits of variability due to random sampling, establishing the reliability of the census method.

DISCUSSION

Estimates by different authors of the number of fibers in the human medullary pyramids or the pyramidal tract at the cervical level are listed in Table 3. The pioneer work of Blocq and Onanoff[8] and Weil and Lassek[9] at the cervical level are not further considered here. The myelin sheath totals of van der Bruggen[10] are so small that some error of method can be assumed, although his data are insufficient to identify it. Verhaart,[11–13] using the Alzheimer-Mann stain, reports the total number of fibers and the range but not the ratio of myelin sheaths to axons. A discrepancy in the range of fibers is apparent in his values for normal human beings, however. The lowest normal count is given as 482,750 fibers[12] and the high-

TABLE 1

CENSUS DATA FOR AXONS AND MYELIN SHEATHS IN HUMAN MEDULLARY PYRAMIDS

No.	*Age*	*Race and Sex**	*Cause of Death*	*Pyramid Area,† mm² (Axons)*	*Total Number of Axons*	*Pyramidal Area,† mm² (Myelin Sheaths)*	*Total Number of Myelin Sheaths*	*Myelin Sheath Total, Axon Total (Per Cent)*
1	19	C, F	Postpartum embolism	10.68	882,700			
2	22	W, M	Acute leukemia	9.68	1,170,600	10.95	940,800	80.4
3	24	W, F	Accidental trauma	11.14	1,205,500	12.87	1,119,600	92.9
4	26	W, M	Metastatic choriocarcinoma	9.19	990,700			
5	28	W, M	Myocardial infarction	11.22	1,092,000	12.50	1,135,900	104.1
6	30	W, M	Diabetic coma	9.68	1,305,100			
7	32	W, F	Tetanus	10.33	1,121,600	12.58	1,063,700	94.8
8	33	W, M	Cardiac arrest	9.09	1,067,600	9.50	956,400	89.6
9	36	W, M	Gunshot	8.29	1,285,500	10.15	1,259,400	98.0
10	42	W, F	Postoperative death—cause unknown	9.49	1,252,500	10.92	1,247,700	99.7
11	48	W, M	Myocardial infarction	9.32	907,400	10.96	936,100	103.2
12	51	W, M	Cardiac arrest	12.01	1,117,800	9.15	995,700	89.1
13	52	W, F	Accidental trauma	8.20	980,900			
14	56	W, M	Myocardial infarction	9.44	906,900			
15	56	W, F	Gunshot	5.98	749,900	6.49	632,800	84.4
16	58	W, M	Abdominal hemorrhage	11.63	1,118,900	14.08	1,045,500	93.4
17	60	W, M	Ruptured abdominal aneurysm	11.79	1,113,200	12.07	1,067,700	95.9
18	60	C, M	Third degree burn	10.29	1,086,100			
19	62	C, M	Acute uremia	10.94	1,238,600			
20	67	W, M	Lobar pneumonia	13.00	1,391,100			
21	68	W, M	Myocardial infarction	7.74	847,600			
Mean	44			9.96 (21 patients)	1,087,200 (21 patients)	11.02 (12 patients)	1,033,400 (12 patients)	93.8 (12 patients)

*M = male F = female C = Negro W = white

†Pyramidal areas uncorrected for shrinkage, which is greatest in the silver impregnated sections because of postfixation in 95% pyridine

TABLE 2
STATISTICAL SUMMARY OF DATA ON FIBER CONTENT OF HUMAN MEDULLARY PYRAMIDS

Item	Number of Specimens	Mean	Standard Deviations ($S_{\bar{X}}$) of Mean and Confidence Interval	Observed Range	Standard Deviation (S_X)	Predicted Range (Mean $\pm$ $t_{.5a} \cdot S_X$)
Axons	21	1,087,200	$S_{\bar{X}a} = 36{,}290$ $P(1{,}011{,}400 < u < 1{,}163{,}000) = 0.95$	749,900 to 1,391,100	166,290	739,700 to 1,434,800
Myelin sheaths	12	1,033,400	$S_{\bar{X}m} = 47{,}918$ $P(928{,}000 < u < 1{,}138{,}800) = 0.95$	632,800 to 1,259,400	165,990	668,200 to 1,389,600
Per cent myelinated fibers	12	93.8%	$S_{\bar{X}m\%} = 2.061$ $P(89.27 < u < 98.33) = 0.95$	80.4% to 104.1%	7.14%	78.1% to 109.5%

TABLE 3
SUMMARY OF COUNTS OF FIBERS IN HUMAN MEDULLARY PYRAMIDS OR PYRAMIDAL TRACT

Level	Number of Specimens	Mean Number of Axons	Range	Mean Number of Myelin Sheaths	Per Cent Myelinated Fibers	Method	Author
Cervical	3	80,000	Not stated			Not stated	Blocq and Onanoff[9]
Cervical	2(?)	250,000	Not stated		About 75%	Bielschowsky and Loyez	Weil and Lassek[9]
Cervical(?)	?			More than 100,000		Weigert	Flechsig, quoted by Weil and Lassek[9]
Medulla	9			150,000		Weigert	van der Bruggen[10]
Medulla	3	1,101,000	1,059,000 to 1,146,000			Davenport	Lassek and Rasmussen[15]
Medulla	2			688,000	61%	Weigert	Lassek[14]
Medulla	Not stated	Not stated	600,000* to 1,000,000			Not stated. Presumably Alzheimer-Mann	Verhaart[12]
Medulla	23	Insufficient data	700,000* to 1,282,000			Alzheimer-Mann	Verhaart[11]
Medulla	3	593,500†	540,920† to 625,520			Alzheimer-Mann and Glees	Verhaart[12]
Medulla	21 (axons) 12 (myelin)	1,087,200	749,900 to 1,391,100	1,033,400	93.8%	Hortega and Spielmeyer	DeMyer (this paper)

*See discussion
†My calculations from Verhaart's data, left pyramid only

est 1,282,000;[11] yet, when he quotes the range, he gives figures of 600,000 to 1,000,000.[13]

The only study comparable to mine is Lassek's[14] using Davenport's silver impregnation and Weigert's stain. Lassek estimated that 61% of the axons from 2 specimens had myelin sheaths. While his mean for the number of axons[15] agrees with mine, my study indicates that around 94% of the axons have myelin sheaths. Lassek states that he was dissatisfied with some of the technical conditions of his study and suggested that his estimate might be too low. In explaining the discrepancy of our results, one can note that Lassek used a paraffin section myelin sheath stain instead of frozen sections. The advantage of

frozen sections over paraffin for optimum study of myelin sheaths is readily recognized in any side-by-side comparison of the 2 methods.

The surprisingly close agreement between the number of axons and myelin sheaths raises the question of whether all the pyramidal axons visible with silver impregnation have myelin sheaths of a size detectable with light microscopy. At least one other group of central fibers, the optic nerve, is thought to be 100% myelinated.[16] The first task is to see whether the difference in the axon and myelin sheath totals results from sampling error. Assuming that the axon and myelin sheath totals are estimates of the same total population, one can apply the Student's t test for paired replicates. The t score is 3.08, indicating that the difference in the 2 estimates would arise only about 1 time in 100 by random fluctuation of sampling error. The counting procedure was next rechecked to determine whether a systematic error had been made in the census. It is of course possible that the difference results from observer error in counting 2 different populations. Other sources of error seem to be that the myelin sheaths cannot be distinguished as clearly as axons, particularly when the fibers orient obliquely as some do in every specimen. In fact, in a few samples in every specimen, the fibers are so oblique that the counts are guesswork. This difficulty is just as likely to increase the total artificially as to decrease it. The ballooning of the sheaths, always seen in paraffin or frozen sections of autopsy material but often labeled a pathologic change, might thin the sheaths enough to escape detection in some places. Since the iron-hematoxylin method requires differentiation, some of the fine fibers might lose the dye, but comparison of apparently optimally differentiated sections with weakly differentiated sections indicates that the thin fibers are equally represented in both. On the other hand, the personal attention of the investigator is necessary to avoid carrying the differentiation beyond the optimum point when some of the fibers will bleach out completely. In most specimens, there seems to be a critical sheath thickness below which no thinner sheaths are seen, rather than a continuum of decreasing size down to invisibility. Whether there are no smaller sheaths or whether this finding indicates the limit of hematoxylin staining cannot be decided with the present analysis. Schmitt,[1] using polarized light, reports that he found all gradations in the size of myelin sheaths. Electron microscopy might show that if sheaths are to be detected by hematoxylin staining and light microscopy, a minimum number of lamellae have to be present. The possibility that electron microscopy might show submicroscopic axons as well as submicroscopic myelin coverings might be another consideration altering the interpretation of this study. To summarize the technical problems, it would appear that any systematic error made in the census of the axons and myelin sheaths would be at the expense of the myelin sheath total.

In an attempt to reach a definite decision about the completeness of myelination as shown by light microscopy, some of the silver impregnated sections were counterstained with Luxol fast blue.[5] Unfortunately, Luxol fast blue, at least when used as a counterstain, shows fewer of the thin myelin sheaths than iron hematoxylin. The hematoxylin method of Baker[17] was not tried because its specificity for myelin sheaths is in doubt.

In lieu of more definitive results, which might be obtained with electron microscopy and other methods not yet applied extensively to the human brain, the best tentative conclusion is that about 6% of the pyramidal fibers are unmyelinated in human beings. In the macaque, 38% of the pyramidal fibers are unmyelinated. Marchi studies of degeneration in the monkey pyramids will be correspondingly less complete than in man.

Although the observed range for the total number of axons in the human pyramids is from 749,900 to 1,391,100, the expected range for 95% of the normal population having no structural disorders of the central nervous system will be from 739,700 to 1,434,800 (mean $\pm$ 2.09 $\times$ standard deviation). Thus, the highest expected values are about double the lowest. Part of the variability of the totals might be the result of fibers branching and continuing in the pyramids as longitudinal fibers. If branching occurs, the fiber totals would be a function of the degree of branching as well as the number of cells of origin of the fibers. It is important to know whether fibers branch and continue a significant distance longitu-

dinally, but no convincing evidence of it could be found in cross and longitudinal sections stained for myelin sheaths and axons. This problem needs to be approached with other methods, perhaps Golgi's.

Since Lassek reports 800,000 fibers in the medullary pyramid[19] of 1 chimpanzee and 748,000 in an adult seal,[20] the range for these animals will overlap the lower limit of the range for man. If one attempts to infer the phylogentic and functional significance of the pyramidal system from fiber totals, one must note that the seal, although highly skilled, has no free digits and makes no discrete digital movements. In the macaque, the highest fiber totals for the pyramid would not be expected to approach 600,000[18] and would not overlap the range for normal human beings.

SUMMARY

As estimated from silver impregnated sections from 21 adult human medullas, the mean number of axons in 1 medullary pyramid is 1,087,200. As estimated from 12 of these medullas, stained with iron hematoxylin, the mean number of myelin sheaths is 1,033,400, or 93.8% of the number of axons. As in all comparisons of this type, the close agreement of the axon and myelin-sheath totals raises the question of whether the difference is the result of a systematic or technical error in the census. The limitations of the study are those of light microscopy and staining methods as well as the problem of observer error.

The observed range of medullary pyramidal axons in this series extends from 749,900 to 1,391,100, while the expected range, including 95% of the population having structurally normal brains, is from 739,700 to 1,434,800. The percentage of myelin sheaths extends from 80.4 to 104.1%. The excess of myelin sheaths over axons in 2 specimens is explained by the variability of the totals owing to random sampling error. The lower limit of the range for axons in man overlaps the range for the chimpanzee and the seal, but the range for man will not overlap the macaque monkey.

The author would like to thank Dr. Orville T. Bailey, for his advice and for the use of his laboratory facilities, and Miss Irene Baird, for her welcome technical assistance.

REFERENCES

1. Schmitt, F., and Geschwind, N.: The axon surface. Progress Biophys. & Biophys. Chem. 8:165, 1957.
2. Fernandez-Moran, H.: The submicroscopic organization of vertebrate nerve fibers. Exp. Cell Res. 3:282, 1952.
3. Lillie, R.: Histopathologic Technique and Practical Histochemistry. New York, The Blakiston Company, Inc., 1954, p. 332.
4. DeMyer, W.: Impregnation of axons and terminal buttons in routine paraffin, or frozen sections of central and peripheral nervous tissue. Am. J. Clin. Path. 29:449, 1958.
5. Magolis, G., and Pickett, J. P.: New applications of the luxol fast blue myelin stain. Lab. Invest. 5:459, 1956.
6. DeMyer, W.: Methods and advantages of random sampling in the census of nerve fibers and other histological elements. Anat. Rec. 125:443, 1956.
7. Nathan, P., and Smith, M.: Spino-cortical fibres in man. J. Neurol. Neurosurg & Psychiat. 18:181, 1955.
8. Blocq, P., and Onanoff, J.: Du nombre comparatif pour les membres supérieurs et inférieurs de l' homme des fibres nerveuses d' origine cérébrale destinées aux mouvements. Gaz. de Hop. 115:248, 1892.
9. Weil, A., and Lassek, A.: Quantitative distribution of pyramidal tract in man. Arch. Neurol. & Psychiat. 22:495, 1929.
10. van der Bruggen, J.: Uber Ersatz der Pyramidenbahnfunktion. Deutsche Ztsch. f. Nervenh. 113:250, 1930.
11. Verhaart, W.: On thick and thin fibers in pyramidal tract. Acta psychiat. et neurol. 22:271, 1947.
12. Verhaart, W.: Hypertrophy of pes pedunculi and pyramid as result of degeneration of contralateral corticofugal fiber tracts. J. Comp. Neurol. 92:1, 1950.
13. Verhaart, W., and Kramer, W.: Uncrossed pyramidal tract. Acta psychiat. et neurol. scandinav. 27:181, 1952.
14. Lassek, A.: Human pyramidal tract, a study of mature, myelinated fibers of pyramid. J. Comp. Neurol. 76:217, 1942.
15. Lassek, A., and Rasmussen, G.: Human pyramidal tract; a fiber and numerical analysis. Arch. Neurol. & Psychiat. 42:872, 1939.
16. Breusch, S., and Arey, L.: The number of myelinated and unmyelinated fibers in the optic nerve of vertebrates. J. Comp. Anat. 77:631, 1942.
17. Brodal, A., and Harrison, R.: Observations on the chemical composition of myelin and the smallest size of myelinated nerve fibers in the central nervous system. Quart. J. Micr. Sc. 89:89, 1948.
18. DeMyer, W., and Russell, J.: The number of axons in the right and left medullary pyramids of Macaca rhesus and the ratio of axons to myelin sheaths. Accepted for publication by Acta Morphol. Neerlando-scandin.
19. Lassek, A., and Wheatley, M.: Pyramidal tract; enumeration of large motor cells of area 4 and axons in pyramids of chimpanzee. J. Comp. Neurol. 82: 299, 1945.
20. Lassek, A., and Karlsberg, P.: The pyramidal tract of an aquatic carnivore (seal). J. Comp. Neurol. 106: 425, 1956.

Steroid suppression of meningeal inflammation caused by Pantopaque

James K. Smith, M.D., and Livia Ross, M.D.

SINCE THE INTRODUCTION of cortisone for treatment of tuberculous meningitis,[1] several experimental and clinical studies have been made which show the effectiveness of steroids in altering the inflammatory response within the subarachnoid space.[2–5] This report is concerned with an application of these agents to the particular variety of arachnoiditis associated with retention of Pantopaque in the subarachnoid space.

METHOD

White rabbits (average weight, 2 kg.) and guinea pigs (average weight, .5 kg.) were used. After ether anesthesia, ethyl iodophenylundecylate (Pantopaque) was injected under the meninges of the animals by a 21-gauge needle through small trephine openings made over the vertex of the skull. Rabbits were injected with 0.5 cc. of the oil, guinea pigs with 0.25 cc. Roentgenograms of the skull confirmed the dye dispersion in the subarachnoid and intraventricular spaces. In a few animals, the effects of mixing blood with Pantopaque and of repeated injections were studied, but histologic examination gave no evidence of any quantitative difference in the arachnoidal response in such animals from responses seen after a solitary injection of Pantopaque. Subarachnoid injection of 0.5 cc. of citrated blood in 1 rabbit showed no arachnoidal reaction three weeks later.

The animals were killed in one to sixty days. Brains were removed, fixed in formalin, x-rayed (Fig. 1), sectioned, and then stained with hematoxylin-eosin or oil red o. and Masson's trichrome stain. After the optimum time and technic for production of a recognizable arachnoidal reaction had been established, another group of animals was similarly prepared except that various types of steroid therapy were administered. The brains of these animals were then compared with those of the control animals.

RESULTS

Rabbits. Of 10 rabbits receiving only Pantopaque, 7 developed a significant arachnoiditis during the survival period of three to sixty days (Fig. 2). In the first week, cellular infiltration was found and, in the second and third weeks, a fibroblastic reaction with thickening of meninges. An animal killed after three days and 2 killed at seven days showed no significant degree of meningeal reaction.

A single injection of 10 mg. of hydrocortisone* was given 2 animals intrathecally concomitant with Pantopaque. The animals, observed for seven and twenty-six days respectively, developed an arachnoidal thickening typical of that seen in untreated animals.

No significant fibrosis or thickening of the pia-arachnoid was seen in 5 rabbits receiving daily intramuscular injections of 25 mg. of cortisone acetate, although patches of round-cell infiltration were occasionally found (Fig. 3).

In 3 rabbits receiving daily parenteral injection of 3 to 5 mg. methylprednisolone acetate† for nine and fourteen days, cellular accumulation of a chronic inflammatory type was noted to be somewhat more extensive in distribution than that in cortisone-treated animals; no significant fibrous thickening was observed. Animals treated with both prednisone and cortisone showed weight loss and emaciation and, toward the end of the observation period, lethargy.

*Hydrocortisone sodium succinate (Solu-Cortef) (Upjohn)
†Medrol acetate (Upjohn)

From the Psychiatry and Neurology Service and Laboratory Service, Veterans Administration Hospital, Oakland, Calif.

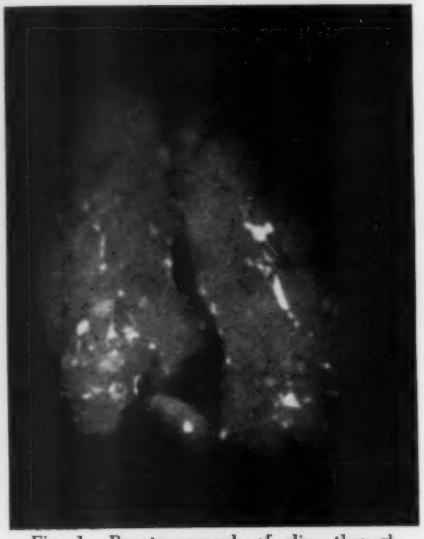


Fig. 1. Roentgenograph of slice through guinea pig brain showing dispersion of Pantopaque beneath the subarachnoid spaces and within the ventricles. (Histologic section from this brain is illustrated in Fig. 5.)

Fig. 2. Rabbit brain twenty-one days after introduction of Pantopaque showing obliteration of the subarachnoid space by proliferation of fibroblasts in the pia-arachnoid membrane. (×100)

Apparently, in the rabbit, cortisone and methylprednisolone inhibit the fibroblastic response of the meninges to Pantopaque – but a single intrathecal injection of hydrocortisone is devoid of such effect.

Guinea pigs. Serving as controls were 8 guinea pigs injected intrathecally with 0.25 cc. of Pantopaque and killed at intervals of one to sixty days. Only 1 – killed after twelve days – had no significant inflammatory process in the meninges; the others all showed an arachnoiditis of varying grades of severity and distribution. The typical finding after one to three days was a well-marked cellular exudate consisting predominantly of polymorphonuclear cells; the typical picture at nineteen to twenty-one days in 4 animals was a fibroblastic proliferation with a tendency to obliteration of sulcal spaces, although without producing an obstructive hydrocephalus in any animal (Fig. 4). Occasionally, some degree of ependymitis was observed.

After receiving the same amount of Pantopaque, 3 guinea pigs were given 0.125 mg.

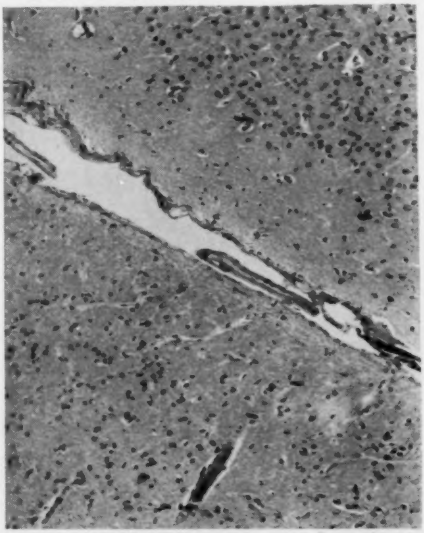


Fig. 3. Rabbit brain twenty-one days after introduction of Pantopaque. Cortisone administered daily (25 mg.). The meninges show minimal reaction and the intercortical space is unobstructed. (×100)

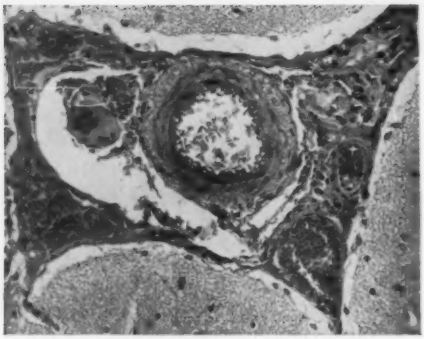


Fig. 4. Guinea pig brain twenty-one days after administration of Pantopaque showing fibrous thickening of pia-arachnoid and tendency to obliteration of the subarachnoid space. (×200)

of methylprednisolone‡ orally every day for fifteen to eighteen days. These 3 animals showed some patchy infiltration of the subarachnoid space with round cells but had no thickening or fibroblastic reaction in the leptomeninges (Fig. 5).

Intramuscular injections of cortisone were given daily to 4 guinea pigs for fifteen to twenty-two days. After administration of the drug for twelve days, it was withheld from 1 animal to see if a delayed fibrous response in the meninges would follow a period of steroid therapy. However, the animal died of purulent meningitis (hemolytic Staphylococcus aureus) three days after cortisone injections were stopped. Probably, low-grade infection in the scalp wound spread to the meninges because of the abrupt withdrawal of cortisone. In another animal, initiation of cortisone was delayed for four days after introduction of Pantopaque; the drug was given intermittently and again discontinued two days before death; at examination an acute purulent meningitis was found with dense accumulations of polymorphs. In 2 remaining guinea pigs given cortisone for sixteen and twenty-two days, the meninges were normal (Fig. 6).

In summary, a significant difference was found between controls and treated animals after fifteen to twenty-five days. All of the control animals showed a fibroblastic proliferation and thickening of the meninges; the animals who received cortisone or methylprednisolone did not. Administration of a single injection of hydrocortisone intrathecally had no observed inhibitory effect.

‡Methylprednisolone (Medrol) (Upjohn)

DISCUSSION

The earliest reports on the clinical use of Pantopaque[6,7] emphasized its safety and the low incidence of unfavorable reactions. Recommendations were made that Pantopaque be removed after myelography; but it was pointed out that when Pantopaque was left within the subarachnoid space, the material was absorbed by the body or, more accurately, became gradually less visible on roentgenograms. In dogs, the absorption was estimated to take place at the rate of 3 cc. annually.

Bering[8] reported no serious complications had been found in 25 patients in whom myelography, with no attempt at removal of the medium, had taken place up to three years previously. He confirmed the gradual decrease of radiopaque material in the subarachnoid space by use of serial roentgenograms. In only 1 patient did he find the Pantopaque immovable under fluoroscope.

However, after use of both lipiodol and Pantopaque, patients occasionally have been reported with arachnoiditis attributable to the irritant action of the foreign material on the meninges. From the pathologic descriptions,

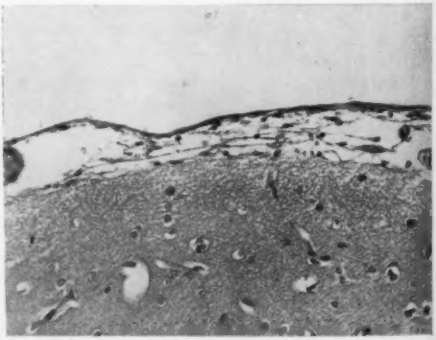


Fig. 5. Guinea pig brain twenty-one days after administration of Pantopaque and daily administration of .125 mg. methylprednisolone. There is a mild reaction with some cellular proliferation and delicate adhesions in the pia-arachnoid. (×200)

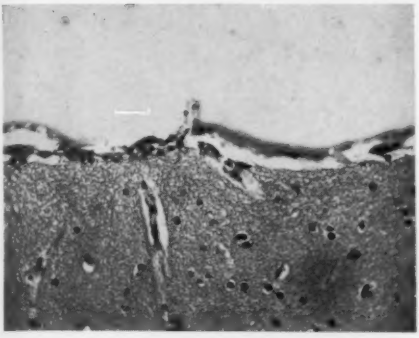


Fig. 6. Guinea pig brain twenty-one days after administration of Pantopaque and daily intramuscular injections of cortisone acetate (12 mg.). The meninges are essentially normal. (×200)

this complication is apparently identical to chronic adhesive arachnoiditis described by Spiller,[9] Horsley,[10] Elkington,[11] and others. A fairly rare condition – often of obscure pathogenesis – chronic adhesive arachnoiditis has been ascribed with some certainty to spinal anesthesia and infection, especially lues. Delayed effects of trauma have also been implicated by some, and at times etiology is unknown. In all the reported cases, certain common features were found: a progressive spread, often in an ascending pattern, of a fibrous thickening of the leptomeninges and eventual formation of cysts; spinal fluid blockage; and, finally, compression of, or cavitation within, the spinal cord.

In addition to the reports of adhesive arachnoiditis after Pantopaque myelography, reports have also appeared of a more immediate meningeal reaction – an aseptic meningitis. Tarlov[12] described a milky opacity of the meninges at laminectomy for a lumbar disk sixty hours after myelography. So far as is known, no serious delayed effects followed.[13] The patient of Bharucha and Desai[14] developed headache, nausea, and fever three hours after a myelogram, and on the following day his spinal fluid contained over 5,000 polymorphonuclear leukocytes and increased proteins. Both clinical symptoms and spinal fluid abnormality cleared within a week.

Peacher and Robertson[15] reported 1 patient with transient signs and symptoms of acute meningitis appearing ten days after a Pantopaque myelogram with incomplete removal of the oil. This patient's spinal fluid yielded 670 lymphocytes. Recovery was complete, and a repeat spinal fluid examination eight months later was normal. The same authors examined the spinal fluid in 20 patients and found increased cells or protein in 12 of them between the second and seventh day after an uncomplicated myelogram. They felt that meningeal reaction of this character was more common after incomplete removal of Pantopaque.

The results of animal experiments, in which Pantopaque is always left in place, vary but are not reassuring from a safety standpoint. Steinhausen[7] and coworkers introduced 3 to 5 cc. of Pantopaque into the cisternal space of dogs and observed transient fever and nuchal rigidity followed by encystment within six weeks and almost complete absorption within a year in "most of the animals." They did not report on the clinical condition of their animals, but histologic sections were said to show a foreign body reaction in the spinal cord.

McLaurin and associates[16] also used the cisternal injection in dogs and found that administration of 3 to 6 cc. of Pantopaque was followed by adhesive arachnoiditis and cavitation of the spinal cord due to constriction of the nutrient arteries.

Differences in species sensitivity and longer contact of the tissues with a proportionately larger dose might be reasons why dogs almost invariably develop this complication which is rare in man. The question of what causes development of a meningeal inflammatory response in some human beings and not in others remains unanswered. Jaeger[17] suggested that the irritant effect of Pantopaque was amplified when it was broken up into small particles – something likely to be caused by the emulsifying effect of blood. Individual idiosyncrasy, dosage, incomplete removal, and sensitization have also been suggested as being responsible. The last point was raised by Luce and associates[18] who reported a local reactivation of an intradermal Pantopaque skin test accompanying an acute aseptic meningitis after myelography. Their first patient developed pain and swelling at the site of a skin test made by intradermal injection of Pantopaque before a my-

elogram. The flare-up of the skin test took place on the ninth day after myelography and was followed a day or two later by headache, chills, stiff neck, and a fever of 102° F. Spinal fluid was turbid; cells increased and protein content was 113 mg. per cent. By the twelfth day, the patient was semistuporous with continued fever, stiff neck, and an absent left patellar reflex; in the next two days he improved and, at the time of the report, had no permanent residuals.

Probably, the great majority of patients who develop acute meningeal inflammation immediately after Pantopaque myelography recover with no further difficulty. However, among this group of acute, benign responses, the occasional, more malignant complication of a late, adhesive arachnoiditis develops. Convincing evidence is lacking, but a link between these two types of reaction is suggested in some of the reported cases. Erickson and Van Baaren's[19] patient had "fever, headache and a flushed face" for two days after a myelogram in which 6 cc. of dye was introduced and not removed. Nine months later the patient had headaches, and six months after this, he was readmitted to the hospital with basilar arachnoiditis – a finding ultimately confirmed by autopsy. Case 1 of Hurteau and associates[20] had fever and severe pain in the back and legs nine days after a myelogram in which Pantopaque was not removed. A laminectomy two years later disclosed a plastic arachnoiditis with cyst formation around the cauda equina.

Since very little, if anything, can be done therapeutically once the end stage of meningeal fibrosis is reached, it would be worth while to check the process at an earlier phase in its development. The studies with animals reported here suggest that corticosteroid drugs provide such a means.

SUMMARY

1] Cortisone and methylprednisolone inhibit the production of fibrous changes in the leptomeninges of animals after subarachnoid introduction of Pantopaque.

2] A review of the literature indicates 2 types of meningeal reaction to Pantopaque: a transient, acute sterile meningitis, and the rarer, late development of adhesive arachnoiditis. In some patients, the possibility of a link between the 2 is suggested.

3] Whenever an acute meningeal reaction is encountered in the early postmyelographic period, particularly in patients in whom Pantopaque was incompletely removed, consideration should be given to a trial course of corticosteroids until clinical and laboratory signs indicate a clearing of the reaction.

It is a pleasure to acknowledge the technical assistance of Mrs. Virginia Smith.

REFERENCES

1. Shane, S. J., Clowater, R. A., and Riley, C.: Treatment of tuberculous meningitis with cortisone and streptomycin. Canad. M. A. J. 67:13, 1952.
2. Feldman, S., Behar, A. J., and Samueloff, M.: Effect of cortisone and hydrocortisone on piaarachnoid adhesions. Arch. Neurol. & Psychiat. 74:681, 1955.
3. Oppenheimer, J. H., and Riester, W. H.: Influence of cortisone on leptomeningeal reaction induced by talc. Proc. Soc. Exper. Biol. & Med. 83:844, 1953.
4. Samueloff, Moni, and Feldman, Shaul: Effect of cortisone acetate administered intracisternally in cats. Arch. Neurol. & Psychiat. 72:599, 1954.
5. Turon, R., Giacardy, R., and Garde, M.: Applications des injections intrarachidiennes d'hydrocortisone en thérapeutique neurologique. Rev. Neurol. 94:304, 1956.
6. Ramsey, G. H., French, J. D., and Strain, W. H.: Iodinated organic compounds as contrast media for radiographic diagnosis; pantopaque myelography. Radiology 43:236, 1944.
7. Steinhausen, T. B., Dungan, C. E., Furst, J. B., Plati, J. T., Smith, S. W., Darling, A. P., Wolcott, E. C., Warren, S. L., and Strain, W. H.: Iodinated organic compounds as contrast media for radiographic diagnosis; experimental and clinical myelography with ethyl iodophenylundecylate (Pantopaque). Radiology 43:230, 1944.
8. Bering, E. A., Jr.: Notes on retention of Pantopaque in subarachnoid space. Am. J. Surg. 80:455, 1950.
9. Spiller, W. G.: Circumscribed serous spinal meningitis. Am. J. Med. Sci. 137:95, 1909.
10. Horsley, Victor: Chronic spinal meningitis. Brit. M. J. 1:513, 1909.
11. Elkington, J.: Meningitis serosa circumscripta spinalis (spinal arachnoiditis). Brain 59:181, 1936.
12. Tarlov, I. M.: Pantopaque meningitis disclosed at operation. J.A.M.A. 129:1014, 1945.
13. Tarlov, I. M.: Personal communication.
14. Bharucha, E. P., and Desai, A. D.: Meningeal irritation following introduction of pantopaque for myelography. Indian J. M. Sc. 8:220, 1954.
15. Peacher, W. G., and Robertson, R. C. L.: Pantopaque myelography; results, comparison of contrast media, and spinal fluid reaction. J. Neurosurg. 2:220, 1945.
16. McLaurin, R. L., Bailey, O. T., Schurr, P. H., and Ingraham, F. D.: Myelomalacia and multiple cavitations of spinal cord secondary to adhesive arachnoiditis. Arch. Path. 57:138, 1954.
17. Jaeger, R.: Irritating effect of iodized vegetable oils on brain and spinal cord when divided into small particles. Arch. Neurol. & Psychiat. 64:715, 1950.
18. Luce, J. C., Leith, W., and Burrage, W. S.: Pantopaque meningitis due to hypersensitivity. Radiology 57:878, 1951.
19. Erickson, T. C., and van Baaren, H. J.: Late meningeal reaction to ethyl iodophenylundecylate used in myelography. J.A.M.A. 153:636, 1953.
20. Hurteau, E. F., Baird, W. C., and Sinclair, E.: Arachnoiditis following use of iodized oil. J. Bone & Joint Surg. 36-A:393, 1954.

Electrogustometric examinations in cerebellopontine tumors and on taste pathways

Bent Krarup, M.D.

SINCE THE TIME of Magendie[1] and Lussana,[2] the problem of the course of the taste pathways from the anterior part of the tongue has given rise to speculations as to the part played by the facial and trigeminal nerves, as well as the anastomoses between them, as conductors of taste impulses.

It is now generally accepted that the most frequent pathway takes the following course—lingual nerve, chorda tympani, facial nerve, intermedius of Wrisberg[3,4]—but many textbooks on anatomy, physiology, and neurology[5–10] describe as many as 5 alternative pathways for taste impulses.

For this reason, neurologists have not paid greater attention to clinical taste examinations; pathologic findings do not yield reliable information on the localization of an eventual lesion of the pathways.

From the relevant literature, the varying opinions on the course of the taste fibers of the fifth and seventh cranial nerves show that the conclusions are very largely based on investigations carried out on patients with nerve lesions caused by neurosurgical operations or injections.

Because several investigators have obtained widely diverging results in examinations of patients of the same category, it was of interest to inquire into the cause of these variances. The explanation would appear to be governed by [1] the close anatomic relationship between the fifth and seventh cranial nerves, making it exceedingly complicated to exclude simultaneous lesions of both nerves and therefore equally difficult to select the clinical material, and [2] the employment of too coarse a technic in the taste examinations. No one has described this problem more clearly than Cushing[11] who, in his book on acoustic tumors, writes: "Unless the tests for the cardinal taste qualities are made with full appreciation of the difficulty of interpreting gustatory impulses, when common sensation of the tongue is affected, errors may easily be made."

It was desirable, therefore, to develop a new technic for gustatory experiments. The method had to be [1] quantitative, thus enabling finer changes to be diagnosed; [2] applicable to a desired area, so that the stimulus could be directed to one part of the tongue without risk of involving other tongue areas; [3] such that a stimulation of the receptors of the sense of taste could not be mistaken for a stimulation of those for common sensation; [4] applicable to as large a number of patients as possible with a minimum demand on the patient's perception and cooperation; and [5] simple and quick, making it practical as a routine clinical investigation.

This paper will describe an electrogustometric technic that appears to fulfill the above demands. It is based on the phenomenon that an electric current passed through the tongue creates a taste impulse of an acid, metallic character. Volta has already demonstrated such a reaction and Hofmann and Bunzel,[12] Hermann,[13] Öhrwall,[14] and Bujas and Chweitzer[15] have all written on this subject.

That a galvanic stimulus can be used in clinical trials is mentioned by DeJong[3] and

From the departments of Otolaryngology and Neurosurgery, The University Clinic Rigshospitalet, Copenhagen, Denmark. This work was supported by grants from the King Christian X Foundation and the P. A. Brandt Foundation.

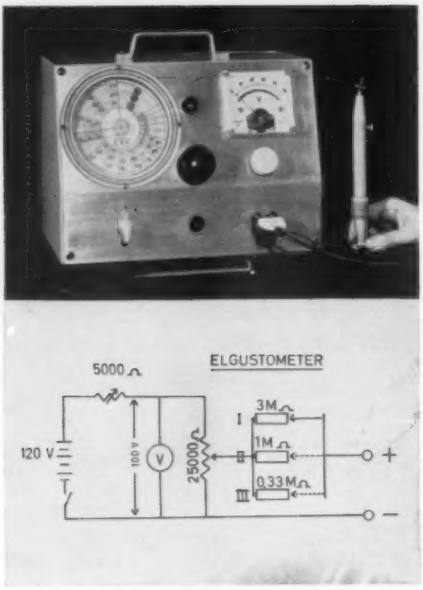


Fig. 1. The Elgustometer[17]

Monrad-Krohn,[16] but the method has not been universally accepted even though it obviously is better than the generally adopted technics using substances representing the taste qualities of sweet, salt, sour, and bitter.

Results obtained with this method will be given, and, on the basis of these results, the problem on the pathways of taste will be discussed.

METHOD

In a previous paper,[17] the construction of an Elgustometer (Fig. 1), an apparatus for galvanic taste stimulation, and the results with normal persons were described. A brief summary of this paper is given here.

The current for the Elgustometer is produced by a dry battery of 120 volts adjusted to 100 volts. The stimulus is regulated with the aid of a Helical potentiometer. The current is led through large resistances to eliminate the great variability of the electrode resistance at various current intensities.

Electrogustometry. The stimulus is recorded in special units called electric gust units (EGU) assessed as follows: The measuring scale, from 5.75 to 300 micro-amp., is divided into 37 divisions with constant percentage variation from unit to unit. The units are marked out on a big circular scale where the hand is coupled to the axis of the potentiometer.

Stainless steel electrodes are used. The tongue is stimulated with the anode. The different electrodes, which have a circular contact area 5 mm. in diameter, are of two types – one has a short shaft for placing on the anterior part of the tongue and the other has a long insulated shaft for posterior placing. The different electrode is mounted in a holder equipped with a silent and, to the patient, invisible switch. The electrode can thus be placed on the tongue partly with and partly without current. The indifferent electrode (cathode) is a disk 4 by 5 cm. which is placed on a piece of gauze moistened with saline solution and attached to the wrist.

For stimulation of the anterior part of the tongue, the electrode is placed about 1.5 cm. from the midline, close to the edge, so that the papillae only are covered by the electrode; for the posterior part, the anode is placed laterally at the base at the circumvallate papillae. The stimulus is applied for one to one and one-half seconds.

A demonstration is first undertaken to enable the patient to sense impression, partly by the application of the electrode minus current and partly with a supraliminal electric stimulus. With this double stimulation, a threshold determination with descending values is carried out to the point where the patient can no longer distinguish between the two applications. The left and right sides of the tongue are taken alternately. Another threshold determination is then carried out with the same double stimulation but this time with increasing values, commencing some EGU below the threshold first found until a difference can again be registered. The final threshold is obtained as the average of the values from further sets of measurements taken as described – the number of measurements depending on the patient's ability to react in threshold determination.

Tests should be given in a quiet room, with only the examiner and patient present. The

tongue should be relaxed and resting on the lower lip; the mouth should be half open. The patient must not be allowed to bite his tongue, and in order to prevent the tongue from drying, the patient should withdraw the tongue into the mouth between each test. Before a test, the electrodes are warmed by friction to feel pleasantly warm on the back of the hand.

The time taken for this test is approximately the same as for another type of threshold determination — a pure tone audiometry.

In this investigation,[17] 140 normal persons ranging from 6 to 80 years of age were tested. On the anterior part of the tongue, the average threshold for the right and left sides varied widely from person to person in all age groups. These values, therefore, hold no greater practical clinical interest. The difference in the thresholds of the right and left sides was of greater clinical value, however, because it was shown that a threshold difference of 3 EGU or more can be considered pathologic. Patients who were unable to recognize an electric stimulus of 37 EGU could, with 98% certainty, be considered to have total ageusia as, normally, only 2% were unable to recognize this stimulus, even though tests with taste solutions showed a sense of taste to be present. Tests carried out on patients with known ageusia have shown that only very rarely has a stimulus of 37 EGU given rise to an irritation of the tongue's receptors for common sensation.

Investigations of the posterior part of the tongue showed that a threshold determination could be carried out in the described manner in 13% of patients. The values were found to be identical from the right and left sides of the tongue. In an additional 36% of normal persons, electric stimulus demonstrated that the threshold obtained was the same from both sides of the tongue without carrying out an exact determination. In the remaining 51%, the investigation was hampered by reflexes, and so on, thus necessitating the use of the semiquantitative test.

Semiquantitative test. This involves the use of 4, 10, and 40% cane sugar, 1, 5, and 10% citric acid, 2.5, 7.5, and 15% sodium chloride, and 0.075, 0.5, and 1% quinine hydrochloride. The solutions were applied to the tongue by means of a pipet, with great care being taken so that these solutions remained on the section of the tongue to be examined. The anterior and posterior parts of the right and left sides of the tongue were tested separately. When considered of particular interest, tests were carried out with saturated solutions of the above mentioned substances.

MATERIAL

Gustatory investigations were carried out on patients after surgery for an acoustic neurinoma in whom [1] the facial nerve had been severed intracranially and [2] sensation in the trigeminal area of the affected side was normal or only slightly reduced. Data were obtained from 10 patients, 8 women and 2 men, aged 42 to 72 years; 1 patient was examined three months after surgery, and the others after two to nine years.

Subject to preoperative gustatory investigations were 11 patients in whom an operation later verified a tumor of the cerebellopontine angle.

RESULTS

The results obtained in the gustatory studies will be recorded as follows:

Electrogustometry
- EGU: Right/left, for example, 9/3
  - $>$ 37 indicates ageusia

Semiquantitative test
- R = L: Thresholds identical on both sides.
- R $>$ L: Threshold higher, that is, lower gustatory sense, on the right than on the left.
- R $<$ L: Threshold higher on the left than on the right.

The investigations on postoperative patients closely corresponded with those of Lewis and Dandy.[18] Without exception, the gustatory sense had been completely lost on the anterior part of the tongue on the side of operation, that is, none of the patients were able to taste the most concentrated solutions in the semiquantitative test nor were they able to recognize an electric stimulus of 37 EGU. On the opposite side, the threshold determinations were normal. This indicates that all gustatory fibers from the anterior part of the tongue pass centrally via the intermedius.

TABLE 1

RESULTS OF PREOPERATIVE OTONEUROLOGICAL EXAMINATIONS IN 11 CASES OF CEREBELLOPONTINE TUMORS (DIAGNOSIS CONFIRMED AT OPERATION)

				Objective Cranial Nerve Involvement (Affected side)									
				Eighth		*Seventh*		*Fifth*		*Ninth*			
*Case**	*Sex*	*Age*	*Side of Lesion*	*Acoustic Function*	*Vestibular Function*	*Motor Function*	*Taste (EGU)*	*Sensation of: Face and tongue*	*Cornea*	*Sensation*	*Taste*	*Porus acusticus internus*	*Remarks*
1	F	41	Left	(+)	0	+	8/17	(+)	(+)	+	R = L	dilated	In Case 2, cyst compressed ninth nerve root.
2	F	48	Left	(+)	(+)	+	17/17	(+)	0	+	25/31	dilated	
3	F	41	Right	(+)	(+)	+	9/3	+	(+)	+	R = L	dilated	
4	F	25	Right	0	0	+	>37/7	+	(+)	+	R = L	normal	
5	M	78	Left	0	0	(+)	27/>37	(+)	(+)	+	R = L	dilated	
6	F	42	Left	0	0	+	23/34	(+)	+	+	R = L	dilated	
7	F	58	Left	0	0	+	10/>37	(+)	(+)	+	R = L	dilated	
8	F	59	Left	0	(+)	(+)	26/36	+	(+)	+	R = L	normal	
9	F	37	Right	(+)	(+)	+	22/22	+	+	+	R = L	dilated	
10	F	35	Left	0	0	+	20/>37	+	(+)	+	R = L	dilated	
11	F	73	Left	0	0	(+)	15/30	+	(+)	+	R = L	normal	

* Cases 1 to 10 were diagnosed as acoustic neurinoma; case 11 as meningioma

\+ Normal function

(+) Impaired function (paresis, hypesthesia, hypacusis)

0 Loss of function (paralysis, anesthesia, anacusis)

Results of the preoperative otoneurologic investigations of patients with cerebellopontine tumors giving the functions of the fifth, seventh, eighth, and ninth cranial nerves are shown in the table. Corneal sensitivity was determined with the aid of the corneasensibilometer constructed by Boberg-Ans.[19] In the table, "impaired hearing function" indicates perceptive hearing loss without recruitment. The vestibular function was determined by the differential caloric test described by Fitzgerald and Hallpike.[20]

All 11 patients suffered from impaired or lost function of the eighth nerve, acoustic as well as vestibular. In addition, 10 patients suffered from gustatory disturbances. In case 2, the sense of taste was normal on the anterior part of the tongue but reduced on the posterior part on the affected side. At operation, the facial nerve was found to be intact in the tumor capsule while a cyst compressed the root of the ninth nerve.

Reduced corneal sensitivity was noted in 9 patients, and reduced sensation in the trigeminal area on the affected side in 5; 3 patients had facial paresis or paralysis.

Apart from the otoneurologic findings, dilation of the porus acusticus internus was found in 8 patients by roentgenographic examination.

In case 3, the semiquantitative test showed the threshold to be equal on both sides, while the EGU thresholds were 9/3.

When a semiquantitative test showed a higher threshold for one taste quality, a proportional increase in all taste qualities was found.

DISCUSSION

Observed results. In agreement with Lewis and Dandy,[18] the writer finds that all taste impulses from the anterior part of the tongue pass centrally via the homolateral intermedius. This is also indicated by the investigations carried out on the preoperative cases.

Gustatory investigations were found to contribute very considerably to the diagnosis of affections in the cerebellopontine angle, in as much as gustatory disturbances were the most frequently occurring symptom, apart from those of the eighth nerve. It would therefore seem natural to assume that an electrogustometric determination can be of considerable diagnostic value in tracing abnormalities in this region.

From the results from taste investigations in earlier works on acoustic tumors, it was not possible to find material sufficiently examined to compare with those reported here. As already mentioned, Cushing,[11] who found gustatory disturbances in 16 of his 30 patients, questioned the reliability of his observations. Lundborg[21] records gustatory disturbances in 36% of his 300 patients belonging to the same category. Gonzales Revilla[22] states that gustatory tests were not carried out in the majority of his 145 patients. He had, however, observed 4 patients with a loss of the sense of taste on the affected side without any other symptoms from the facial nerve, "indicating that taste fibres might be affected at times much earlier than the motor fibres." This is in accordance with the results given here, where there were motor facial disturbances in only 2 of 8 patients suffering from acoustic neurinoma with gustatory disturbances of the anterior part of the tongue. That the intermedius is affected before the motor section of the facial nerve may be due to the fact that the intermedius, as its name implies, lies between the acoustic nerve and the main section of the facial nerve. Thus, it may be possible to make a differential diagnosis between the acoustic neurinomas and the meningiomas in the cerebellopontine angle, because the sense of taste would, in the former, be affected before the facial motor fibers and, in the latter, the opposite would be the case.

Course of the taste pathways. It has already been shown that taste fibers from the anterior part of the tongue follow the chorda tympani in all cases.[23] It can now also be shown that they continue along the facial nerve and intermedius. The question must now be raised: Why do so many investigators indicate that the trigeminal nerve also plays an important part as a pathway of taste? In an attempt to answer this question, the claims of the most frequently quoted writers and those of the author will be compared.

It appears that the results obtained by these writers were based on investigations of patients who had lost their sense of taste and at the same time suffered anesthesia in the

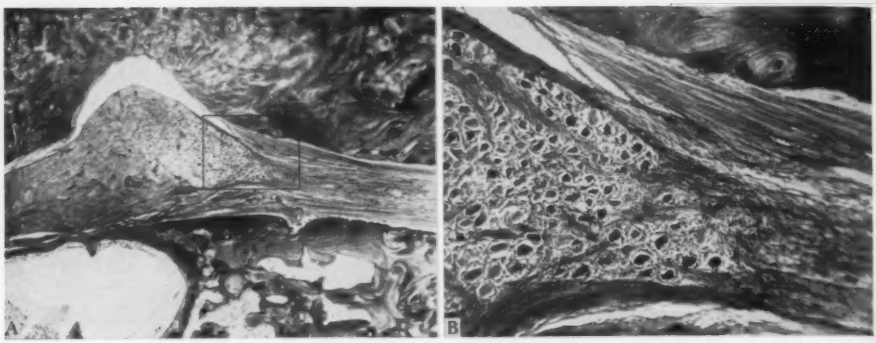


Fig. 2. *(A)* Section through the geniculate ganglion of the facial nerve at the entrance of the greater superficial petrosal nerve. *(B)* Detail of *(A)* in higher magnification

corresponding trigeminal area after the gasserian ganglion or the third trigeminal branch had been damaged through neurosurgical procedures. Apparently, objections can be made partly to the choice of clinical material and partly to the technic employed for the gustatory investigations. These sources of errors will be commented on separately below:

• Unobserved lesions of the geniculate ganglion via hiatus fallopii or of the chorda tympani resulting from operation on the gasserian ganglion or injection of alcohol in the ganglion or the third trigeminal branch

It has already been shown by Dixon[24] that an extirpation of the gasserian ganglion can easily lead to a damaging effect on the facial nerve via the fallopian canal when the dura is stripped off. In a great number of patients given alcohol injections in the gasserian ganglion or the third branch, Harris[25] found unimpaired taste sense in only about 10% and concluded that taste impulses must be conducted by the trigeminal nerve.

Carmichael and Woollard[26] found a loss of sense of taste in 10 out of 17 patients of the same category but demonstrated, by injecting colored gelatin into cadavers, how easily the injected substance passed along the central trigeminal root to the posterior fossa and through the fallopian canal to the geniculate ganglion. That the taste fibers of this ganglion can be affected without the motor function of the facial nerve being involved is explained by the writers by assuming that the cells of the ganglion are more vulnerable than the motor fibers.

In order to obtain proof of this theory, the writer, at the Temporal Bone Laboratory of the ENT department, has examined serial sections of the geniculate ganglion to ascertain the relationship between the motor fibers and the ganglion cells at the entrance of the fallopian canal. It will be seen from Figure 2 that the canal with the greater superficial petrosal nerve emerges at the point where the ganglion cells are situated so that these are first affected by penetrating injected fluid and therefore provide protection for the motor fibers, which, furthermore, are protected by their myelin sheaths.

Injections into the third trigeminal branch or electrocoagulation of the nerve with Kirschner's technic may give rise to a damage of the chorda tympani, particularly so if the chorda takes a steep course. Since all taste impulses travel through the chorda tympani, this would naturally lead to a loss of the sense of taste on the corresponding area of the tongue. The writer has personally examined 4 patients of this category; 3 possessed a normal sense of taste while the fourth suffered from total ageusia.

Theories which are based on investigations of patients of these categories should therefore be accepted with great reservations.

• Application of too highly concentrated solutions in gustatory investigations

The employment of too concentrated solutions of the taste substances may give misleading information, thereby creating not only a taste stimulation but also an irritation of

the receptors for common sensation. Thus, an anesthesia can simulate an ageusia.

Schwartz and Weddell[27] have reported a case of lost sense of taste in a patient in whom a resection of the trigeminal root had been performed and in whom anesthesia developed without motor facial symptoms. The workers used crystalline substances moistened with water whereby very high concentrations of the substances are obtained.

Rowbotham,[28] who also uses moistened crystalline substances, found reduced or eliminated sense of taste in 34 out of 40 patients on whom a root resection had been performed. Some examples from my personal experience follow:

Case A. This woman of 47 years of age had a later verified neurinoma of the left gasserian ganglion. Anesthesia presented in the left trigeminal area, but facial function was normal.

Gustatory tests:

EGU: 8/4

Semiquantitative: R = L

Saturated solutions: These solutions are tasted best on the right, or unaffected, side, an indication of reduced sense of taste on the opposite side.

Case B. This 48-year-old man had had resection of the right trigeminal root fourteen years previously. Total anesthesia of the right side was present. Facial function was normal.

Gustatory tests:

EGU: 7/17

Semiquantitative: R = L

Saturated solutions: As in case A, a quicker reaction was noted on the nonoperated side.

On the side of anesthesia, normal taste reactions were elicited with both electrogustometry and the semiquantitative tests. That the taste thresholds are lower on the operated side is explained by the fact that the electric stimulus is more easily registered when the application of the current free electrode is not sensed at all because of total anesthesia. Where only concentrated solutions were used in the tests, the immediate conclusion drawn would be that the sense of taste was considerably reduced on the side of the lesion, since these solutions would have an irritating effect dominating the reaction of the taste buds.

For these reasons, investigations carried out under such conditions should be regarded with reservation.

• Unobserved lesions of the chorda tympani.

Since all taste fibers from the corresponding section of the tongue follow the chorda tympani,[23] any lesion would inevitably cause gustatory disturbances. If an otologic examination of a patient is unsatisfactory, it is possible that a chorda lesion is responsible for a lost, or reduced, sense of taste. This is illustrated by the following two case studies.

Case C. A 57-year-old woman had had a resection of the right trigeminal root four years previously. Postoperative anesthesia in the corresponding trigeminal area resulted, but facial function was normal.

Gustatory tests:

EGU: 24/18

Semiquantitative: R > L

Otoscopic examination revealed cicatrices in the right tympanic membrane, particularly near the malleolar folds.

It cannot be excluded that an earlier infection of the middle ear may have damaged the chorda tympani.

Case D. This man of 74 years of age was suffering from trigeminal neuralgia.

Preoperative examination revealed normal trigeminal and facial function. Otoscopic examination was also normal.

Gustatory test:

EGU: 15/15 (normal)

A decompression of the left gasserian ganglion was performed by the temporal route. After the operation, the patient complained of a subjective sensation of the loss of taste on the side of operation. Examination at this time revealed hypesthesia in the left trigeminal area and normal function of the facial nerve.

Gustatory test:

EGU: 17/> 37 (ageusia on the left side)

When the patient complained of a blocked feeling in the left ear, further otoscopy was carried out since it seemed most likely to be due to ear wax or coagulated blood in the external auditory canal. This was not the case, but a hematotympanum on the left side was found.

The observed hematotympanum indicates an accidental fracture of the temporal bone due to the craniotomy, giving rise to a possible damage or severance of the chorda tympani. This would explain the left-sided ageusia.

From these observations, it would appear that Cushing[29] was correct in stating that an extirpation of the gasserian ganglion does not

result in a permanent loss of taste. He observed that there could be a temporary reduction in the sense of taste for a short period after the operation but connected this with the degeneration of the fibers of the lingual nerve, which takes place because of toxic or other causes of trophic disturbances. For this latter reason, minor disturbances in the sense of taste may be conceded when electrogustometry is used as this technic is more sensitive than the methods hitherto employed. An example is shown.

Case E. This 51-year-old man had had a decompression of the left trigeminal root performed three years earlier. Anesthesia of the left trigeminal area developed. Facial function was normal.

Gustatory test:

EGU: 10/15

Slight reduction of sense of taste on the side of operation was the result.

These examples show the necessity of extreme care in the choice of clinical material and technic of examination when gustatory investigations carried out on patients suffering from trigeminal disorders are used for tracing the course of the taste pathways. In all, 37 patients with trigeminal disorders have been examined at the University Clinic by the technic described in this paper. The patients with one-sided hypesthesia or anesthesia and pathologic differences of the taste thresholds on the right and left sides could all be placed in one of the above mentioned categories. Consequently, the examinations have not given support to the theory that the trigeminal nerve should have a function as a conductor of taste impulses.

CONCLUSION AND SUMMARY

A description of an electrogustometric technic for the determination of taste thresholds is given. The method would appear to be reliable and well suited for clinical use. The results are more reliable and more accurate than those obtained from a semiquantitative method involving the use of test solutions. A purely qualitative technic using highly concentrated test solutions obviously can only give rough and, in many cases, misleading estimates of gustatory changes. The writer, therefore, is of the opinion that this method should no longer have its place in neurologic diagnostics when technics for investigations of the functions of the other cranial nerves have been improved to existing standards.

Gustatory investigations belong to the otoneurologist since a careful otoscopy is essential for a well performed gustatory investigation.

On the basis of the clinical findings of tumors in the cerebellopontine angle and the subsequent discussion on earlier works on the course of the taste pathways, it would seem reasonable to assume that the taste fibers from the anterior part of the tongue, in all cases, take the following course: lingual nerve, chorda tympani, facial nerve, intermedius of Wrisberg. This is in complete agreement with the observations made by Cushing and Dandy fifty-five and twenty-eight years ago, respectively.

REFERENCES

1. Magendie, F.: Précis elémentaire de physiologie, ed. 2, vol. 1. Paris, Méquignon-Marvis, 1825, p. 143.
2. Lussana, A.: Recherches experimentales et observations pathologiques sur les nerfs du gout. Archives de Physiologie normale et pathologique 2:20, 1869.
3. DeJong, R. N.: The Neurological Examination. New York, Paul B. Hoeber, Inc., 1950, p. 225, 230.
4. Brodal, A.: Neurological Anatomy in Relation to Clinical Medicine. Oxford, Oxford University Press, 1948, p. 239.
5. Ranson, S. W.: The Anatomy of the Nervous System, ed. 5. Philadelphia and London, W. B. Saunders Co., 1936, p. 174.
6. Spalteholz, W.: Hand-Atlas of Human Anatomy (translated from German). London and Philadelphia, J. B. Lippincott, 1943, p. 723.
7. Mitchell, G. A. G.: Anatomy of the Autonomic Nervous System. Edinburgh, E. & S. Livingstone Ltd., 1953, p. 163.
8. Brain, W. R.: Diseases of the Nervous System, ed. 2. London and New York, Oxford University Press, 1945, p. 194.
9. Wright, S., Maizels, M., and Jepson, J. B.: Applied Physiology, ed. 9. London and New York, Oxford University Press, 1952, p. 581.
10. Grinker, R. R., and Bucy, P. C.: Neurology, ed. 4. Springfield, Charles C Thomas, 1949, p. 221.
11. Cushing, H.: Tumors of the Nervus Acusticus and the Syndrome of the Cerebellopontine Angle. Philadelphia and London, W. B. Saunders Co., 1917, p. 167.
12. Hofmann, F., and Bunzel, R.: Untersuchungen über den elektrischen Geschmack. Arch. ges. Physiol. 66: 215, 1897.
13. Hermann, L.: Beiträge zur Kenntnis des electrischen Geschmacks. Arch. ges. Physiol. 49:519, 1891.
14. Ohrwall, H.: Untersuchungen über den Geschmacksinn. Skandinav. Arch. für Physiologie 2:1, 1891.
15. Bujas, Z., and Chweitzer, A.: Contribution à l'étude du gout dit électrique. L'année psychologique 35: 147, 1934.
16. Monrad-Krohn, G. H.: Clinical Examination of the Nervous System, ed. 10. London, H. K. Lewis & Co., Ltd., 1954, p. 76.
17. Krarup, B.: Electro-gustometry: a method for clinical taste examinations. Acta oto-laryng. 49:294, 1958.

18. Lewis, D., and Dandy, W. E.: Course of nerve fibers transmitting sensation of taste. Arch. Surg. 21: 249, 1930.
19. Boberg-Ans, J.: Experience in clinical examination of corneal sensitivity. Brit. J. Ophth. 39:705, 1955.
20. Fitzgerald, G., and Hallpike, C. S.: Studies in human vestibular function; observations on directional preponderance ("Nystagmusbereitschaft") of caloric nystagmus resulting from brain lesions. Brain 65:115, 1942.
21. Lundborg, T.: Diagnostic problems concerning acoustic tumors; study of 300 verified cases and Békésy audiogram in differential diagnosis. Acta oto-laryng. Supp. 99:1, 1952.
22. Revilla, A. G.: Neurinomas of the cerebellopontile recess; a clinical study of 160 cases, including operative mortality and end results. Bull. Johns Hopkins Hosp. 80:254, 1947.
23. Krarup, B.: Taste fibres and the chorda tympani. Acta oto-laryng. Supp. 140:201, 1958.
24. Dixon, A. F.: On the Course of the Taste Fibres. Edinburgh M. J., 1 (new series):395, 1897.
25. Harris, W.: Neuritis and Neuralgia. London, Oxford University Press, 1926, p. 247-281.
26. Carmichael, E. A., and Woollard, H. H.: Some observations on fifth and seventh cranial nerves. Brain 56:109, 1933.
27. Schwartz, H. G., and Weddell, G.: Observations on pathways transmitting sensation of taste. Brain 61: 99, 1938.
28. Rowbotham, G. F.: Observations on effects of trigeminal denervation. Brain 62:364, 1939.
29. Cushing, H.: Taste fibres and their independence of the N. trigeminus; deductions from 13 cases of gasserian ganglion extirpation. Bull. Johns Hopkins Hosp. 14:71, 1903.

Hypersensitivity of in vitro myasthenic muscle to d-tubocurarine

John B. Dillon, M.D., and Phiroze B. Sabawala, M.D.

In 1934, Dale and Feldberg[1] described the role played by acetylcholine in the transmission of the nerve impulse across the neuromuscular junction. In this same year, Walker[2] described the efficacy of physostigmine in the treatment of a patient with myasthenia gravis.

By correlating these two discoveries, Hamill and Walker[3] suggested that myasthenia gravis was caused by some defect in transmission at the neuromuscular junction. This opinion differs in many respects from the theory that myasthenia is caused by a circulating toxin elaborated by an abnormal thymus gland which is based on older concepts – beginning with Weigert's study[4] on the association of thymomas and myasthenia. The latter theory is supported by some experimental work[5] and the statistical evidence of the improvement in the condition of these patients after thymectomy.[6] Arguments for and against these two views can be found in Osserman's review of the subject.[7]

Apart from the well-known hypersensitivity of myasthenic patients to d-tubocurarine,[8] some apparently normal subjects are very sensitive to the drug. Gray and Halton[9] observed complete paralysis after injecting only 2 mg. of d-tubocurarine chloride into an unanesthetized subject. The close resemblance between the paralysis of myasthenia and that produced by curare was noted by Walker,[2] who later provided indirect evidence to show that active contractions of muscles in myasthenic patients caused the liberation of a curare-like substance into the circulation.[10]

We have been conducting experiments on human intercostal muscle to test its possible use as an efficient pharmacologic preparation. Although work on this problem is still in progress, the dramatic results obtained with the first muscle specimen secured from a myasthenic patient seem definite enough to warrant this preliminary report.

METHODS

Strips of external intercostal muscle were taken during thoracotomy for various diseases of the chest from nonmyasthenic patients and from a 6-year-old myasthenic child on whom a thymectomy was performed.

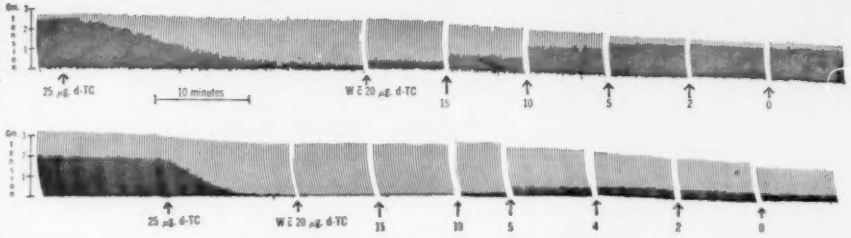
Aside from the fact that in the infant intercostal muscle is short and difficult to work with, the age of the patient from whom the specimen is removed does not in any way affect the performance of the muscle. However, the nonmyasthenic specimen included in the present study was removed from a 5-year-old girl with a patent ductus arteriosus so that the age difference would not be too great in the two records shown.

These strips of muscle, intact from origin to insertion, were then carefully dissected parallel to the long axes of the muscle fibers.[11] The resulting preparations were about 1.5 cm. long under optimum tension, 1 to 2 mm. thick, and weighed 25 to 27 mg. when wet. The muscle bundle was then attached to a platinum electrode holder and immersed in a bath containing a modified Krebs saline.[11] The saline was equilibrated by gassing with a mixture of 95% O_2 and 5% CO_2 for thirty minutes; temperature was kept at 37° C. by warm water circulating through a jacket surrounding the bath.

The muscle was stimulated at the rate of one stimulus every ten seconds alternately, directly and indirectly through the nerve axons lying within the tissue.[12] A clipped con-

From the Department of Surgery, Division of Anesthesiology, Medical Center of the University of California at Los Angeles.

This work was supported by grant-in-aid No. B-1203 from the U.S. Public Health Service.



Response of in vitro intercostal muscle strips to gradually decreasing doses of d-tubocurarine chloride. Samples taken from (A) nonmyasthenic 5-year-old girl with patent ductus arteriosus and (B) myasthenic 6-year-old girl with enlarged thymus. D-tubocurarine was added in the dose and at the points shown. Each interval in the record represents a pause of one hour in otherwise continuous recordings. Throughout these records, the smaller twitch is the one evoked by indirect stimulation.

denser discharge stimulator[11] provided the stimuli which were adjusted to a slightly supermaximal value. The muscle contractions resulting from this stimulation were recorded by a Statham strain gauge and an Offner dynograph. A dose of 25 μg. of d-tubocurarine chloride added to the bathing medium is sufficient to produce a complete neuromuscular block, after which the physiologic saline is washed out with 50 cc. of fresh saline containing the next lower dose of d-tubocurarine. Equilibrium between drug and receptor site was obtained in about forty-five minutes.[13]

RESULTS

Figure A is a record representing steady, partial, neuromuscular blocks produced by the addition of varying doses of d-tubocurarine chloride to the bathing fluid. The behavior of nonmyasthenic intercostal muscle under the conditions of this experiment have been previously reported.[11] Figure B is the record of a similar experiment performed with a sample of intercostal muscle removed from a myasthenic patient. The obvious differences between these two records are:

1] The large, apparent block seen in the short control record before curare is added to the fluid bathing the myasthenic preparation is a peculiarity of this particular muscle and can be reversed by the addition of prostigmin.

2] With a dose of curare, 25 μg. per 50 cc. Krebs saline, sufficient to produce a 100% neuromuscular block in the nonmyasthenic muscle, the onset and completion of the block in the case of the myasthenic muscle are 3 times as rapid.

3] At a dose level of between 10 and 15 μg. per 50 cc. Krebs, nonmyasthenic muscle is only 50% paralyzed, while the response to indirect stimulation in the case of myasthenic muscle has just reappeared at this dose.

4] Even with repeated washing, the height of contraction evoked by nerve stimulation is difficult to restore to its control height in myasthenic muscle.

No definite conclusions can be drawn from the results of this single experiment, but, apparently, in myasthenia gravis, the defect in neuromuscular transmission lies primarily at the junction itself and is not due to some toxin in the patient's circulation; if present, the toxin would have been removed by repeated washing with the artificial medium used in these experiments.

SUMMARY

The response of an in vitro preparation of intercostal muscle from a myasthenic patient after the preparation was subjected to varying doses of d-tubocurarine chloride has been described. A comparison is made between this response and that shown by nonmyasthenic muscle. The results seem to show that the defect in neuromuscular transmission in myasthenic muscle is probably due to some change at the neuromuscular junction itself.

ACKNOWLEDGMENTS

The authors gratefully acknowledge the valuable technical assistance of Mr. Floyd Oppenheimer and the cooperation of the staff of the Department of Surgery, Medical Center, University of California at Los Angeles.

REFERENCES

1. Dale, H. H., and Feldberg, W.: Chemical transmission at motor nerve endings in voluntary muscle. J. Physiol. 81:39P, 1934.
2. Walker, M. B.: Treatment of myasthenia gravis with physostigmine. Lancet 1:1200, 1934.
3. Hamill, P., and Walker, M. B.: The action of "prostigmin" (Roche) in neuro-muscular disorders. J. Physiol. 84:36P, 1935.
4. Weigert, C.: Pathologisch-anatomischer Beitrag zur Erb'schen Krankheit. Neurol. Zbl. 20:597, 1901.
5. Wilson, A., and Stoner, H. B.: Myasthenia gravis: consideration of its causation in study of 14 cases. Quart. J. Med. 13:1, 1944.
6. Keynes, G.: Surgery of thymus gland. Brit. J. Surg. 33:201, 1946.
7. Osserman, K. E.: Studies in myasthenia gravis. New York State J. M. 56:2512, 1956.
8. Bennett, A. E., and Cash, P. T.: Myasthenia gravis; curare sensitivity; new diagnostic test and approach to causation. Arch. Neurol. & Psychiat. 49:537, 1943.
9. Gray, T. C., and Halton, J.: Idiosyncrasy to d-tubocurarine chloride. Brit. M. J. 1:784, 1948.
10. Walker, M. B.: Myasthenia gravis: case in which fatigue of forearm muscles could induce paralysis of extra-ocular muscles. Proc. Roy. Soc. Med. 31:722, 1938.
11. Creese, R., Dillon, J. B., Marshall, J., Sabawala, P. B., Schneider, D. J., Taylor, D. B., and Zinn, D. E.: The effect of neuromuscular blocking agents on isolated human intercostal muscles. J. Pharmacol. & Exper. Therap. 119:485, 1957.
12. Rushton, W. A. H.: Excitable substances in nerve-muscle complex. J. Physiol. 70:317, 1930.
13. Holmes, P. E. B., Jenden, D. J., and Taylor, D. B.: Analysis of mode of action of curare on neuromuscular transmission; effect of temperature changes. J. Pharmacol. & Exper. Therap. 103:382, 1951.

Akinesia in Parkinson's disease

Robert S. Schwab, M.D.,* Albert C. England, M.D.,† and Elizabeth Peterson

In 3 of 5 patients described by Parkinson[1] in 1817, weakness and profound muscular fatigue were conspicuous symptoms in addition to tremor and rigidity. In 1925, Kinnier Wilson[2] further emphasized this striking difficulty in motor strength and endurance in patients with parkinsonism but could not specifically correlate it with either rigidity or amount of tremor present. He commented, "It seems as if the patient does not care to continue the task or put an adequate amount of effort into a particular movement."

No pathologic evidence exists that any break occurs in the continuity of the voluntary motor system – such as is found in pyramidal tract disease caused by stroke or in lower motor neurons affected by neuritis or poliomyelitis. Strong motivation in the ordinary patient with Parkinson's disease will produce a perfectly normal level of muscle power. Therefore, the word paralysis in the term paralysis agitans is not really justified.

For this study of akinesia, we have developed a special, accurate ergograph in which the first dorsal interosseous muscle in the hand is specifically studied. The rest of the hand and wrist are held in a fixed position. The patient can, by using this muscle, abduct his index finger toward his thumb, pulling a lever on which is an adjustable weight. The movement of the finger is registered on moving paper as an ergogram (Fig. 1). A half-hour after a voluntary muscle ergogram has been obtained, the apparatus with the identical settings of finger, wrist, and arm positioning points is again connected to the patient. Then, the electrode from a Batrow muscle stimulator is placed over the motor point of the first dorsal interosseous muscle and the electronic[3] ergogram produced as previously described.

Figure 2 shows examples on the left, under A, of a normal ergogram obtained by an apparatus with a 10-oz. weight on the bar. The rested subject was able to perform this amount of work indefinitely; amplitude was even and sustained. On the right of A, the same muscle is being stimulated and is producing the electronic ergogram. When the electrode approximates 2 cm. in diameter over the motor point of the muscle, the amount of weight the normal muscle can pull is only 4 oz.; the muscle does not have more than 60% of the amplitude of the voluntary ergogram. This is because the electrical stimulation is limited to the area mentioned – 2 cm. of muscle – while, with voluntary use, the entire muscle and the adductor pollicis muscle are utilized. Therefore, with the voluntary ergogram versus the electronic ergogram of the same muscle, we have a ratio of $\frac{\text{Voluntary}}{\text{Electronic}}$ of approximately 3 to 1. This ratio varies in some individuals – according to skill in locating the motor point – and may be 3 to 1, 2 to 1, or even 6 to 1.

Using a 10-oz. weight, a patient with Parkinson's disease tires easily (Fig. 2B, left). However, at the arrow, the patient is told to make a strong effort and immediately returns the ergogram to normal levels for 1 contraction only; the ergogram then falls back to greatly diminished value. When a patient with Parkinson's disease uses a 4-oz. weight, repetition of the test an hour later shows little improvement. Again on urging the patient, the ergogram reaches toward normal values as indicated under the arrow. Figure 2B, left, represents 4 oz. In Figure 2C, the other arm of the same patient, as shown in B, is tested demonstrating the impairment with the 10-oz. or the 4-oz.

From the Neurological Service of the Massachusetts General Hospital and Harvard Medical School, Boston.
*Assistant Clinical Professor of Neurology, Harvard Medical School; Neurologist, Massachusetts General Hospital, Boston.
†Instructor in Neurology, Harvard Medical School, Boston.
Read at the tenth annual meeting of the American Academy of Neurology, Philadelphia, April 25, 1958.
Supported by the Allen P. and Josephine B. Green Foundation, Mexico, Mo.

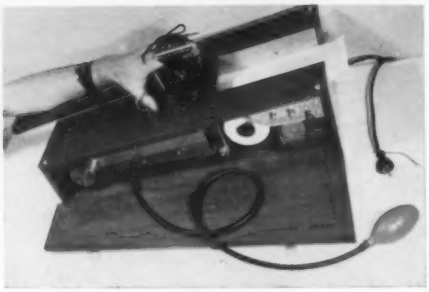


Fig. 1A. The ergographic apparatus used in this study. The weight on the scale beam is shown on the left. The completed ergogram is shown on the paper, and the pen can also be seen. It is possible, also, with this unit to switch from the weight-recording ergograph to an ergograph driven by squeezing a rubber bulb. (Apparatus manufactured by Massey Dickinson Company, Watertown, Mass.)

Fig. 1B. The Batrow electrode stimulator as it is placed over the first dorsal interosseous muscle for production of the electronic ergogram. (Stimulator and electrode manufactured by Batrow Laboratories, Inc., Branford, Conn.)

weight and the ability of the patient to increase the amplitude voluntarily on urging under the arrow. Note that the direction of the ergogram is reversed when the hand is changed. In Figure 2D, another patient with this symptom is shown who was unable to move the 10-oz. weight and even with the 2-oz. weight has irregular movements. Note that in Figure 2 the electronic ergograms of the normal A and the Parkinson B and C are essentially the same. The severe case D has a somewhat diminished electronic ergogram, but it is clear that in all 3 of these Parkinson cases the electronic ergogram is much better than the voluntary.

Numerous instances are recorded of a sudden emergency, for instance, fire, which motivates a woman bedridden with Parkinson's disease to spring to her feet, snatch a sleeping child from its crib, and run downstairs to safety. When such extreme stimulation no longer exists, the patient quickly reverts to invalidism. One can speculate on reality rejections, reactive depressive states, diffuse motivation dullness, and other schizoid or withdrawal conditions, but the preservation of keen sensory perception and alert and sensitive awareness in many patients with pseudoweakness symptoms makes these emotional explanations untenable.

DEFINITION

Rather than dyskinesia, we have selected the word *akinesia* to cover these motor deficits, because dyskinesia nearly always means involuntary muscle activity such as tremor, athetosis, and so on – symptoms which are not true components of the symptom complex we are describing. Weakness, a term A. E. Walker prefers, is usually associated with impairment in performance at the muscle. Usually, reaching a maximum level of motor power is not difficult, and neither emergency stimuli nor motivational changes can augment this by more than 10 to 15% (Fig. 3).

Akinesia includes the following subjective complaints:

1] The patient's almost constant awareness of fatigue in a limb used for ordinary, repetitive motor acts such as walking, writing, or brushing the teeth. In extreme forms, the patient reports a deep, ill-defined ache in the affected limb.

2] A peculiar, rapid exhaustion of amplitude which occurs in such movements; zero values are reached at low levels of energy expenditure.

3] Difficulty experienced by the patient in shifting from one motor contraction pattern to another. This freezing is frustrating and causes strong emotional stress.

4] An almost pathologic reduction that occurs in the patient's achievement goals; the simplest tasks are not completed and others never begun. Accompanying this apathy is an almost complete dependence on other people.

Objectively, akinesia includes:

1] Reduction in level of most motor tasks.

2] Pathologic or extreme fall-off from reduced to lower levels (Fig. 4).

3] Inability to complete actions.

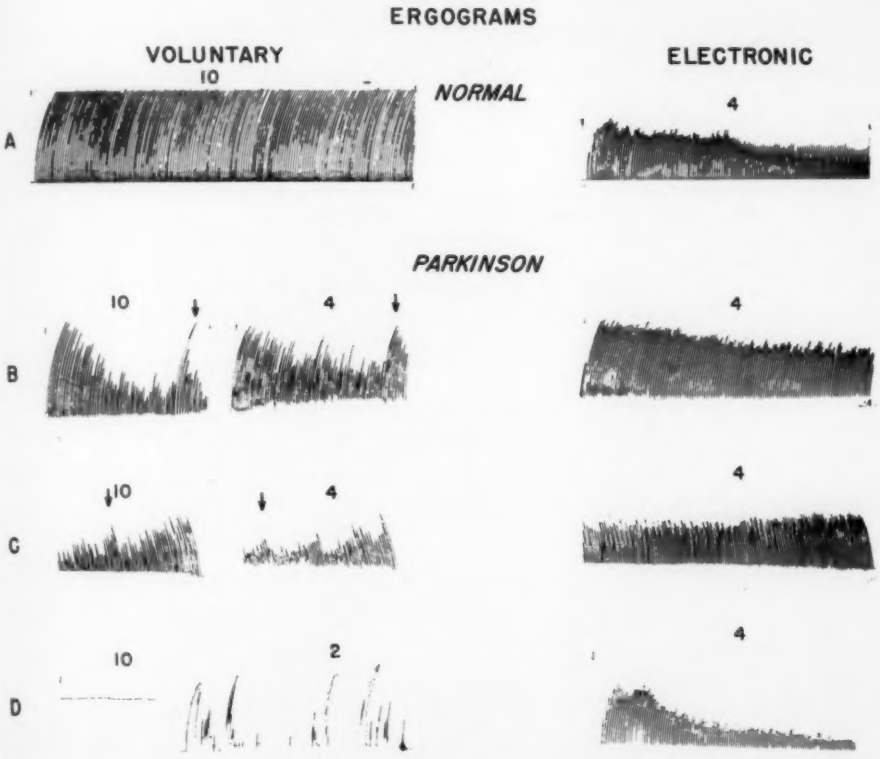


Fig. 2. This shows examples on the left, under A, of the normal voluntary ergogram obtained by this apparatus with a 10-oz. weight on the bar. The subject is not fatigued and is able to do this amount of work indefinitely. Amplitude is even and sustained. On the right of A, stimulation of the same muscle produces the electronic ergogram. At B, left, is the left hand of a patient with Parkinson's disease, with voluntary ergogram with 10-oz. weight showing the wrist fatigue curve. At the arrow, the patient is urged to make a vigorous squeeze, which he does with normal amplitude, which is about 5 times the amplitude of the previous squeeze. To the right of this curve, under No. 4, is a repeat ergogram with a 4-oz. weight, showing little benefit or change from reducing the amount of work done and the same increase in amplitude with urging under the arrow. To the right of B is the electronic ergogram in this same limb, which is a normal curve and shows about 3 times the work done as the voluntary. Under C is the record of the same patient with his right arm being tested, showing reduced amplitude. A rise with urging is demonstrated in both cases. On the right of C is the electronic ergogram of the right arm which is again normal and perhaps 8 times the output of the voluntary. Note that the change from left to right involves reversing the direction of the ergogram. Under D is the record of a more severe case with inability to move the 10-oz. weight, marked difficulty with the 2-oz. weight, and a relatively normal electronic ergogram on the right.

4] Difficulty in releasing active muscles.

5] Extreme loss in ability to start or sustain one act when another must be done.

6] Loss of many cooperative or associated motor acts producing mechanical inefficiency of an extreme degree.

7] An almost constant ability to reach normal levels briefly under sudden motivation (Fig. 2). The ratio of high motivation is 3 to 5 times that of ordinary motivation, while other ordinary kinds of motor deficits, or those in healthy subjects, respond to an urge for a better contraction by only a 10 to 15% increase in amplitude.

8] Electronic ergograms in healthy individuals run one-third of the maximum of voluntary ergograms because only a part of the muscle available for action is stimulated (Fig. 2). In akinesia, the ratio is reversed; the voluntary ergogram is one-half to one-tenth of the near normal electronic ergogram.

CLINICAL CORRELATION

In approximately 50 patients with Parkinson's disease, we have no relationship in the fatigue appearance of the voluntary ergograms to the presence of tremor in the involved limb or to the amount of resting tremor. For example, some patients with 4-plus tremor in the left hand had an almost normal voluntary ergogram.

A 63-year-old chemistry professor with tremor in his left arm and hand had no symptoms on the right side. He was able to do all his motor tasks without impairment, including the voluntary ergogram. His principal difficulty was the emotional embarrassment the tremor caused him in public, particularly when he was in front of his class.

Rigidity, on the other hand, has been uniformly associated with some impairment in the voluntary ergogram; a drop in the amplitude occurs early, suggesting muscular fatigue. Slight involvement with rigidity to the level of 1-plus, for example, is not associated with evidence of akinesia; extreme forms of rigidity, such as 3-plus or 4-plus, show some evidence of akinesia as described. This is particularly noticeable in patients who have unilateral symptoms – such as rigidity on one side, but not on the other. The voluntary ergogram shows a normal appearance on the uninvolved

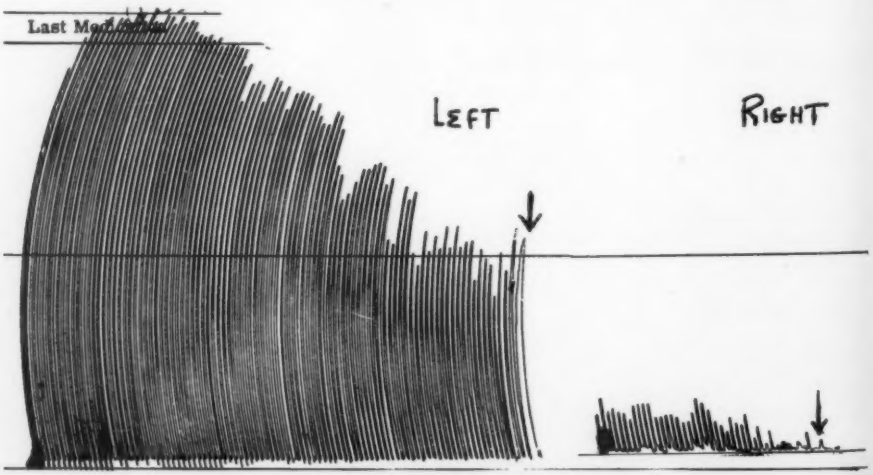


Fig. 3. A voluntary bulb ergogram of a patient with right-sided hemiplegia. A powerful curve is found on the healthy left side. While fatigued, the patient is unable to give an extra-large contraction. This is the normal finding. In the right, hemiplegic ergogram, the curve is one-twentieth of normal and, at the end, when the patient is very tired, his effort to give an additional squeeze produces only a tiny excursion, a result which is not found in akinesia. The arrows indicate in each curve the squeeze under increased motivation.

side and a pathologic appearance on the involved side. Extreme loss of facial expression either bilaterally or on one side or loss of associated movements is not correlated with akinesia. On the other hand, when amplitude and intelligibility of speech are not maintained and the patient has difficulty swallowing or chewing food, we always find evidence of akinesia in the arms and hands. Such patients almost always have bilateral involvement, which seems to be one of the specific correlations with akinesia.

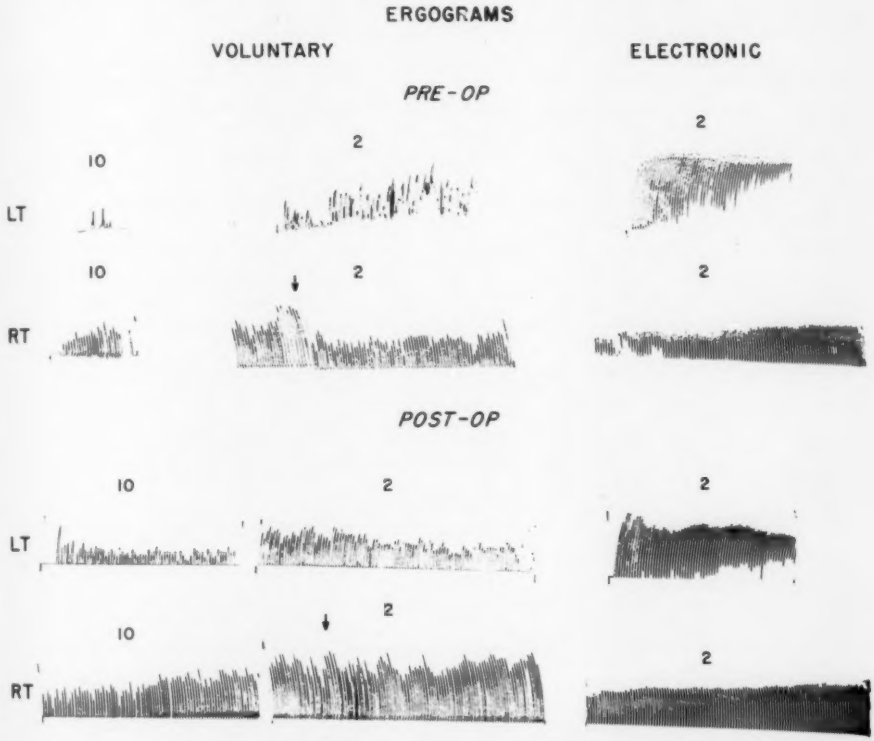


Fig. 4. Examples of voluntary and electronic ergograms in a 40-year-old patient with Parkinson's disease without tremor who had moderate rigidity on the left side and minimal rigidity on the right side. She had moderate akinesia in the involved left side and slight akinesia in the right side. On the preoperative study, she could not move the 10-oz. weight significantly with the left hand and was only able to move the 2-oz. weight irregularly with the left hand. The electronic ergogram on the left side is essentially normal. On the right side, the 2-oz. voluntary test was slightly larger than the 2-oz. electronic ergogram. In the middle of the 2-oz. test, the patient was able, with an effort, to double her initial squeeze, as indicated by the arrow. A globus pallidus lesion was made on this patient on the right side which completely eliminated the rigidity on the left. Marked clinical improvement occurred as well, but it is not difficult to show in the three months' postoperative study that akinesia is still present. Note that there is marked improvement in the patient's ability with voluntary ergogram on both the left and right sides with the 10-oz. weight. The 2-oz. weight is still done better with the electronic ergogram on left than the voluntary. On the right, unoperated side, the postoperative ergogram shows a voluntary 2-oz. curve that is slightly greater than the electronic curve; this is a slight shift toward a normal pattern. Both voluntary and electronic right ergographs begin recording on the right side and proceed to the left; the left-sided ones move in the other direction.

In a recent study,[4] we have found that approximately 40% of the electroencephalograms in Parkinson's disease are abnormal. The more specific abnormalities, where the rhythms show much 5 to 6 per second activity, are associated with this syndrome of akinesia. When extreme akinesia is found, one can expect a bilaterally abnormal electroencephalogram which suggests diffuse brain disorder. Further confirmation is shown by a pneumoencephalogram of lateral ventricular enlargement—especially of the third ventricle—which is correlated with the presence of akinesia in patients with Parkinson's disease. Of patients with extreme akinesia, 80% have shown this abnormality in air studies.

A further correlation with akinesia is the slow, insidious development of sensitivity to the belladonna drugs such as stramonium, hyoscine, and Artane. Sensitivity to sedative drugs, such as barbiturates, also develops.

Another symptom of akinesia is the patient's tendency to lose his normal alertness; often he is content to remain inactive for hours at a time. Perhaps this inactivity is correlated with the rigidity previously mentioned. However, in some patients with severe akinesia, rigidity and tremor may hardly exist. Patients may sleep fifteen hours a day and fall asleep in the presence of interesting friends or while watching exciting entertainment on television. For example, even when the patient's possible admission to a nursing home is being discussed, he may fall asleep in the presence of his relatives and his physician. Indifference is not the correct explanation for this condition.

The most disturbing clinical correlation with akinesia is the development of the patient's invalidism far beyond the degree of rigidity and tremor present. Patients with advanced akinesia are almost completely helpless and dependent on others. We can state unequivocally that the presence of akinesia is a very serious symptom in a patient with Parkinson's disease.

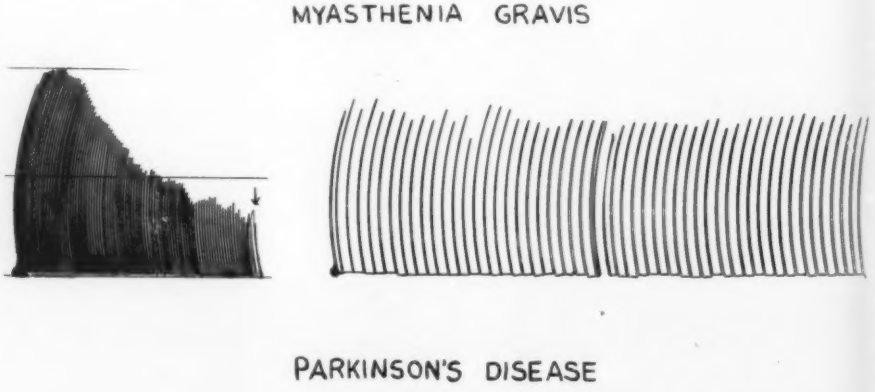


Fig. 5. *Top left:* The voluntary bulb ergogram of a myasthenic patient who is well adjusted to medicine. A fatigue curve is produced with the right hand, and, at the end, the patient is only able to bring it a few per cent above the other level. This is indicated at the arrow. *Lower left:* Bulb ergogram of a patient with extreme parkinsonism who is able, on request, to bring the height to 4 times the value of the last squeeze. On the right side is a bulb ergogram in which the four seconds' rest interval is allowed to take place between each squeeze; other squeezes were once a second. Note in the case of the myasthenia patient that this rest period enables the patient to continue without fatigue all the way across the paper. In the case of the patient with parkinsonism and akinesia, however, this rest period is of no value. The same type of fatigue curve occurs.

TREATMENT

Most of the medications effective against tremor and rigidity do not help akinesia. In fact, usual doses of such medications seem to increase symptoms. We have found that analeptics, such as the amphetamines or Metrazol, strychnine, and ephedrine, seem to help this disabling symptom. However, improvement, even when large amounts of these analeptics are combined in a shotgun prescription, is pitifully slight. Of course, the patient's family is grateful for even 5 to 10% reduction in the invalid's dependence.

Physiotherapy may help this symptom slightly.

The most important asset to a patient with extreme akinesia is an understanding, energetic, helpful spouse. Without him or her, the patient is neglected, develops bedsores, loses weight because of improper diet, and usually has to be sent to a hospital or nursing home. However, if akinetic symptoms increase, such a move becomes mandatory – no matter how helpful the patient's family.

When akinesia is unilateral, mild, and related to the amount of rigidity present, surgical intervention in the globus pallidus improves but does not eliminate it (Fig. 4). When the symptoms of akinesia are extreme and symptoms are present bilaterally, akinesia usually increases – even if surgical intervention is only unilateral. In our opinion, operation is contraindicated. For example, one akinetic patient lost unilateral rigidity after operation but was unable to use the affected arm and leg as efficiently as before.

CASE HISTORIES

Case 1. A 63-year-old, retired business man had influenza in France in 1918. In 1946, he began to drag his left foot and noticed a loss of left-sided dexterity while playing golf and tennis. In 1947, Parkinson's disease was tentatively diagnosed. However, the patient did not have tremor and had only slight rigidity in the left side. Pneumoencephalograms and electroencephalograms were normal.

Since 1947, loss of function, dexterity, initiation, and performance has progressed in the patient's left side, and his right side has slowly become involved. He did not have tremor. In 1952, the patient was almost unable to work and showed increasing sensitivity to belladonna compounds. Freezing began to appear in 1954, and, in 1957, the patient was unable to do anything for himself.

Psychologic tests show that the patient has lost 50% of his intellectual capacity. Exacerbation of dullness and freezing is caused by administration of one-fourth an ordinary dose of Pagitane. Analeptics are the only drugs the patient can tolerate, but benefit is slight. The patient's electroencephalogram is now diffusely abnormal at 6 to 7 per second activity bilaterally.

Ergograms on both sides vary from one-fifth to one-half of normal production and can be modified consistently by increasing the motivation level. On Sept. 18, 1957, the last ergogram showed more reduction on the right side where tremor was zero and rigidity minus. At the end of the test, when the height of contraction was 3 to 4 mm., a specifically induced contraction raised the height to 4 cm. In spite of this, the electronic ergogram was very close to normal.

Case 2. A 57-year-old man in good health who never had had influenza first noticed a loss of coordination and dexterity in his left hand when playing tennis. A year later, the leg became involved with wrist difficulty and pain. In 1954, the patient's left hand trembled when he was under strain. In 1956, the patient's condition had progressed, and his right hand was awkward and clumsy. In the hospital, the patient was benefited by administration of small amounts of Artane. By 1957, the patient had 2-plus tremor and rigidity on the left side and 1-plus on the right; his total score was 38%.

In December 1957, the patient was readmitted to the hospital for relief of left-sided rigidity by a globus pallidectomy. Preoperatively, his left ergogram was approximately 50% of normal, while the right ergogram was 75% of normal. The patient's intellectual capacity was considerably reduced. The electroencephalogram was borderline; the preoperative ventriculogram showed slight dilation of the third ventricle.

At operation, a lesion in the right globus pallidus was made which completely eliminated the left-sided tremor and rigidity. However, the patient remained confused and dull, and his convalescence lasted five weeks.

When the patient was seen three months after surgery, rigidity on the left side was minus; the right side remained the same. The voluntary ergogram on both left and right sides was slightly less than before operation. Electronic ergograms were also the same on both sides and close to normal values. No preoperative electronic ergograms were done on this patient.

Other signs of akinesia have increased, in spite of reduction of rigidity and tremor in this patient.

Case 3. In 1948, a 67-year-old physiology professor who had never had influenza or encephalitis began to notice slight fatigue, particularly in the left arm and hand. In 1951, her left hand began to tremble. In 1952, the patient lost dexterity in her right hand and became awkward and slow. She also had difficulty concentrating and remembering. In 1954, the patient retired.

At onset of the disease, slight rigidity tremor was found on the left side. Ergograms were reduced and fell off to amplitudes of one-twenty-fifth of normal. However, the patient was always able to give a final squeeze 10 to 15 times the value of the last one.

The patient was sensitive to medicine and was administered 1 tablet of Cogentin a day. She became emotionally unstable and had extreme difficulty in organizing household tasks. The electronic ergogram was considerably higher than the voluntary ergogram; single finger movements were invariably easier to do than bulb squeezing.

DISCUSSION

A healthy subject can squeeze a rubber bulb on a recording ergograph at 1 per second without fatigue, loss of amplitude, or rise of the baseline. The healthy subject can also squeeze the bulb with little loss in amplitude while his other hand is doing another voluntary task. In our experience with healthy individuals, evidence of impairment does not occur, even when motivation is not high.

On the other hand, if a patient has myasthenia gravis with a specific, pathologic exhaustion of transmission at the myoneural junction, premature loss of amplitude and real fatigue result. Many situations produce reactions similar to extreme akinesia in parkinsonism. The fatigue curves of the ergogram in myasthenia gravis do resemble those seen in the akinesia of parkinsonism.

However, if the myasthenic patient has an adequate rest and then squeezes the bulb at every fourth bell, pathologic fatigue vanishes and amplitude is down only 5%. In the akinetic patient, the spreading of the ergographic task to every fourth bell produces the same fall-off as before, indicating that the patient is not benefited by rest.

If a patient with extreme akinesia, whose fatigue curve drops rapidly, is asked at the end of the test to give a strong, single contraction, the patient can produce one almost normal contraction that may be 2 to 3 times higher than the initial squeeze and 10 to 15 times higher than the last squeeze. After a rest period, if a reward such as a silver medicine box is dangled in front of the patient's eyes, if motivation is high, and the nurse, doctor, and relatives shout their encouragement, the patient can double the size of his curve, although irregularities and some fall-off still occur. In other types of motor impairment, such as myasthenia gravis under such artificial motivational stimuli as described above, these patients will overinnervate many muscles and show a large autonomic response but fail to increase the ergographic performance by more than 5%.

Even extreme, 4-plus tremor does not interfere with performance of muscular work of this kind, according to our grading. Apparently, rigidity seems to play a certain specific factor so that 3-plus to 4-plus rigidity, for example, will reduce the ergographic performance approximately 25%. When this rigidity is relieved by medication or operation, the curve reaches normal again. When the ergographic curve is greatly reduced and extreme rigidity is absent, evidence of akinesia is always present. Perhaps 1 in 5 akinetic patients has difficulty letting the spring or weight return the pen to the baseline. This causes a small rise of the baseline.

In general, as akinesia progresses and disability becomes more evident in patients, ability to perform with the ergograph diminishes.

SUMMARY

The symptoms of akinesia in patients with Parkinson's disease have been described as motor impairment resembling fatigue that is not related to tremor. As shown by the ergogram, the fall-off can be temporarily altered by a brief rise in motivation. It is related to abnormal electroencephalograms; cortical atrophy by air studies; sensitivity to belladonna and sedative drugs; and loss of intellectual ability, alertness, and motivational drive. It is made worse by the usual medications and sedatives and by neurosurgical intervention. It is helped slightly by analeptics, physiotherapy, and devoted and understanding families.

REFERENCES

1. Parkinson, J.: An essay on the shaking palsy. London, 1817. Sherwood, Neely and Jones.
2. Wilson, S. A. K.: The Croonian Lectures on some disorders of motility and of muscle tone with special reference to the corpus striatum. Lancet 2:1-10, 53-62, 1925.
3. Schwab, R. S., and De Lorme, T. L.: Ergograms, electronic and voluntary, in the various neurological disorders. Med. contemp. 73:2, 1955.
4. Schwab, R. S., England, A. C., and Peterson, E.: Comparison of electroencephalographic findings in Parkinson's disease in 1937 with 1957. Electroencephalog. & Clin. Neurophysiol. 10:3, 186, 1958.

Use of human tissue culture cells to attempt isolation of virus from multiple sclerosis patients

George E. Seiden, M.D.

PREVIOUS ATTEMPTS[1] to isolate and cultivate an infectious agent from multiple sclerosis patients by inoculation of blood and spinal fluid into growth media and laboratory animals have been unsuccessful. The recent development of stable cell line tissue culture technics made possible a new attempt to isolate such an agent by inoculation of specimens into mammalian and human cells maintained in tissue culture.

MATERIAL AND METHOD

Spinal fluid and blood for inoculation were obtained from 8 chronically ill chair or bed patients from the neurologic service of the Philadelphia General Hospital and 34 patients from the neurologic services of the University of Pennsylvania, Bryn Mawr, and Philadelphia Veterans Administration hospitals. Longstanding multiple sclerosis had been diagnosed in the former group, acute relapses of the disease in the latter.

Spinal fluid was collected from all patients by lumbar tap and in most instances inoculated into cells within four hours or shell frozen and stored at Dry-Ice temperatures before inoculation. Approximately one-fifth of the specimens were stored at 3° C. for twelve to thirty-six hours before inoculation or shell freezing.

Heparinized venous blood specimens were collected in 19 patients with clinically diagnosed acute relapses and serum specimens obtained in an additional 5. The specimens were either inoculated into cells or stored at Dry-Ice temperature after removal of the red cells by centrifugation.

Hela, amnion, HEP II, M, conjunctiva, and pituitary cells were used for inoculation. Amnion cells were obtained by trypsinization of placentas freshly obtained from the obstetric delivery rooms; HEP II, conjunctiva, and hela cells from stable cell lines carried at the Wistar Institute; M cells, an apparently stable cell line, from the white matter of brain of Macacus rhesus;[2,3] and pituitary cells by trypsinization of pituitary tissue of a 6-month-old infant who died of traumatic causes.

In the case of hela, conjunctiva, M, and pituitary cells, monolayer tubes were prepared before inoculation. The cells were washed with a maintenance medium consisting of 2% horse serum in Eagles and then overlayered with 2 cc. of this medium containing 100 units of penicillin, 50 μg. of streptomycin, and 30 units of mycostatin per cubic centimeter. Tubes were inoculated in duplicate with 0.2 cc. of spinal fluid or serum and incubated at 37° C. Control tubes were inoculated with equivalent amounts of normal spinal fluid or serum and with saline. The cells were inspected over a period of five days or until cytotoxic changes were observed.

At the end of five days, or upon the appearance of cytotoxic changes, 0.5-cc. aliquots from these tubes were reinoculated into freshly prepared monolayer tubes. In all, 5 serial passages were made over a period of about three to four weeks.

Amnion and HEP cells were treated in a slightly different fashion. The originally inoculated monolayer of amnion tubes was maintained by fresh changes of maintenance medium every four days for three weeks. HEP cells were planted in milk dilution bottles before inoculation and maintained after inoculation by change of maintenance medium consisting of 5% calf serum in Eagles over a period of two to three months.

From the Wistar Institute, Philadelphia.
Supported by Grant 161 from the National Multiple Sclerosis Society.
Present address: Albert Einstein College of Medicine, New York City 61.

During the period of serial passage or maintenance, cells were examined microscopically and stained with Giemsa after methyl alcohol fixation.

After the fifth serial passage in M, hela, pituitary, and conjunctiva cells and after conclusion of maintenance periods of amnion and HEP cells, remaining cells were scraped from glass, suspended in the residual maintenance medium, and frozen to be tested serologically as antigen against possible antibodies in pooled multiple sclerosis patient serum.

The complement fixation test was employed. Heat-inactivated (57° C. for thirty minutes) tissue culture cells and supernatant were used as antigen and heat-inactivated convalescent multiple sclerosis serum pooled from 40 patients as the source of antibody. Antigen and antibody were permitted to react with 2 full units of complement at 6° C. for eighteen hours and at 37° C. for one-half hour. The degree of fixation was determined by observing the degree of hemolysis resulting upon incubation of the residual complement with 2 units of hemolysin and sheep red blood cells.

Complement and hemolysin were titrated freshly before each test. Antigen and antibody were tested for hemolytic and anticomplementary activities. Control complement fixation tests, using egg white antigen and syphilitic serum, were also performed.

RESULTS

No passable cytotoxic agent was discovered in any of the sera, bloods, or spinal fluids tested. Fresh heparinized plasma from some patients was observed to be cytotoxic on first inoculation into hela and HEP cells, but this effect could not be passed. Fresh specimens of heparinized plasma from normal laboratory workers and from resident physicians were also observed to produce this nontransmissible effect in varying degrees. It was determined that heparin in saline was not cytotoxic, nor could serum without heparin produce the effect when taken from the same individuals whose heparinized plasma was cytotoxic. The cytotoxic effect consisted largely of clumping of cells, causing their detachment from glass.

The serologic tests revealed no significant degree of complement fixation with any of the tissue culture fluid supernatants used. None of the cell lines employed was anticomplementary after heat inactivation.

DISCUSSION

It was not possible to isolate a transmissible cytotoxic agent from the blood or spinal fluid of patients with acute and chronic multiple sclerosis by means of human tissue culture methods. This study does not rule out the viral or infectious etiology of multiple sclerosis because [1] although human cells were used in all but one case (M cells are derived from monkey brain), they were not of human central nervous system origin and could conceivably be improper host cells for such an experiment; [2] viruses are notoriously difficult to isolate from spinal fluid and blood in known virus diseases of the central nervous system, so that it may be necessary to use tissue from central nervous system lesions as a possible source of virus; [3] specimens may have been taken from patients at improper times during the course of the illness, the proper time to search perhaps being before the development of neurologic lesions; and [4] the virus may be capable of latent existence in cells both in and out of the body. It was for this last reason that serologic tests were employed to search for a proliferating antigen in the absence of tissue culture cytotoxic effects. The complement fixation test did not reveal the proliferation of such an antigen; more sensitive serologic tests may have been necessary.

SUMMARY

Human cell tissue cultures and the tissue culture of the central nervous system of monkeys failed to isolate a transmissible cytotoxic agent from the blood and spinal fluid of patients with acute and chronic multiple sclerosis. Further, it was not possible to determine the proliferation of antigen by serologic testing of tissue cultures against serum pooled from patients with multiple sclerosis.

REFERENCES

1. McAlpine, D., Compston, N., and Lumsden, C. E.: Multiple Sclerosis. Baltimore, Williams & Wilkins; and Edinburgh, Livingstone, 1955.
2. Seiden, G. E.: Morphologic and biochemical studies of a tissue culture cell line derived from the white matter of the brain of Macacus rhesus. Part I, to be published in Experimental Cell Research.
3. Korey, S. R.: Morphologic and biochemical studies of a tissue culture cell line derived from the white matter of the brain of Macacus rhesus. Part II, to be published in Experimental Cell Research.

ULTRAN® helps you to restore assurance

In a wide range of diseases which are primarily organic, apprehension, anxiety, and tension may obstruct recovery. In such cases, adjunctive therapy with Ultran as an aid to your reassurance will often equip the patient better **for a smooth return to normal living.**

Ultran (1) allays apprehension and anxiety, (2) relieves neuromuscular tension, and (3) enhances the effectiveness of analgesic therapy. It is well tolerated, notably safe, and chemically unique.

Supplied in Pulvules® of 300 mg. (usually 1 t.i.d.) and scored tablets of 200 mg. (usually 1 q.i.d.).

Ultran® (phenaglycodol, Lilly)

ELI LILLY AND COMPANY • INDIANAPOLIS 6, INDIANA, U.S.A.

974002

NEW

GRASS

MODEL IV

SIXTEEN CHANNEL ELECTROENCEPHALOGRAPH

- Same accuracy, dependability and operating convenience which have made Model III internationally famous.
- For applications in clinical and research Electroencephalography and Electrocorticography requiring up to sixteen channels.
- Can be supplied with any number of channels when eventual expansion to more than eight is contemplated.
- Up to 36-point electrode selection available.
- Model IV has the same dimensions as Model III except for an increase in depth of four inches.
- Model IV does not replace or make obsolete Model III, which is produced for applications requiring a maximum of eight channels.

GRASS INSTRUMENT COMPANY

101 OLD COLONY AVENUE - QUINCY, MASS.

for those with

PARKINSONISM

"... in our experience procyclidine (Kemadrin) proved a worthy addition to the therapy of parkinsonism, because it afforded relief to many patients who had failed to respond to other drugs. It exerts an action against all symptoms of parkinsonism ... hence it may be employed as the basic drug in commencing treatment with new cases."

Zier, A. and Doshay, L. J.: Procyclidine Hydrochloride (Kemadrin) Treatment of Parkinsonism in 108 Patients, Neurology (July) 1957.

"... in our series of 30 severe Parkinsonism sufferers, 21 obtained moderate to good relief with the use of this new agent, Kemadrin, in combination with other drugs."

Lerner, P. F.: Kemadrin, a New Drug for Treatment of Parkinsonian Disease, J. Nerv. & Ment. Dis. 123:79 (Jan.) 1956.

Smoother activity,
and *brighter expression*
with **'KEMADRIN'**®

Also indicated for the treatment of drug-induced symptoms resembling parkinsonism, developing during treatment of mental patients.

'KEMADRIN' brand Procyclidine Hydrochloride
Tablets of 5 mg., scored. Bottles of 100 and 1,000.

BURROUGHS WELLCOME & CO. (U.S.A.) INC., Tuckahoe, N. Y.

Before the fact . . . closed-ward management

SPARINE quickly controls the excitement and hostility of acute and chronic psychoses. As a practical adjunct to formal psychiatric measures, SPARINE simplifies care, facilitates accessibility, speeds social rehabilitation.

SPARINE gives prompt control by intravenous injection and effective maintenance by the intramuscular or oral route. It is well tolerated in all three methods of administration.

Comprehensive literature supplied on request

Sparine®
HYDROCHLORIDE
Promazine Hydrochloride

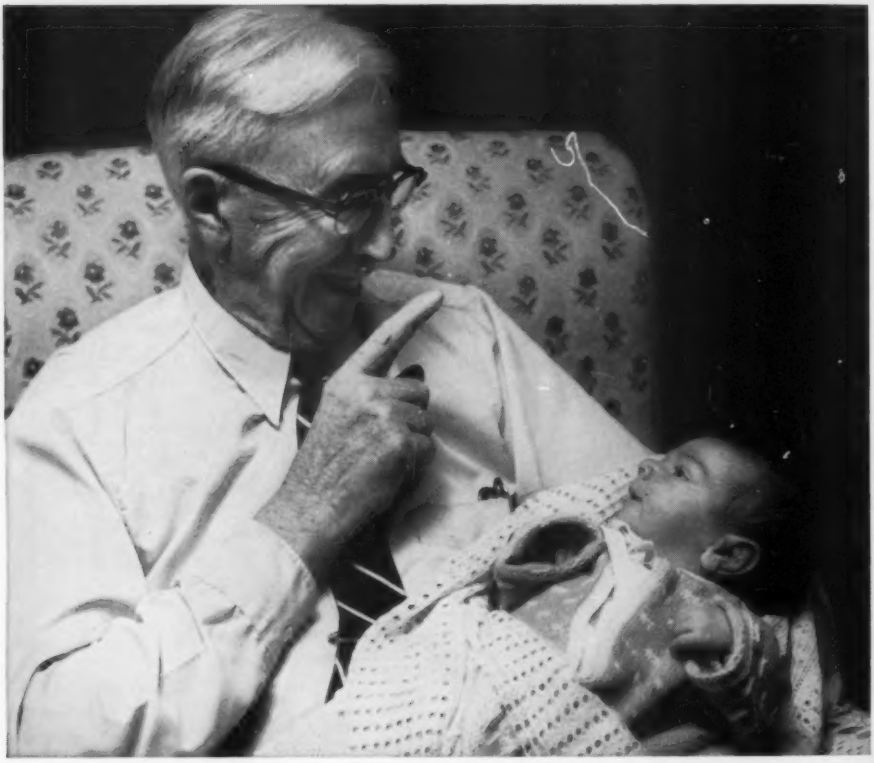
INJECTION TABLETS SYRUP

Wyeth®
Philadelphia 1, Pa.

EQUANIL®
Meprobamate, Wyeth
PHENERGAN® HCl
Promethazine HCl, Wyeth
SPARINE HCl
Promazine HCl, Wyeth

A Wyeth normotropic drug for nearly every patient under stress

HELP US KEEP THE THINGS WORTH KEEPING



Each of us wants peace for his own precious reasons. But peace costs money. Money for strength to keep the peace. Money for science and education to make peace lasting. And money saved by individuals to keep our economy strong. Each Bond you buy helps provide this money—helps strengthen America's Peace Power. Are you buying *enough?*

HELP STRENGTHEN AMERICA'S PEACE POWER

BUY U. S. SAVINGS BONDS

The U. S. Government does not pay for this advertising. The Treasury Department thanks, for their patriotic donation, The Advertising Council and this magazine.

NEWSLETTER

AMERICAN ACADEMY OF NEUROLOGY

Francis M. Forster, President
Augustus S. Rose, President-Elect
A. L. Sahs, Vice-President
Joseph M. Foley, Secretary
Charles Van Buskirk, Treasurer

JANUARY 1959

Special Review Courses To Open 1959 Academy Meeting

The AAN tradition of opening their annual meetings with a series of intensive special review courses will be followed again at the Academy's Eleventh Annual Meeting in the Statler-Hilton, Los Angeles (April 12-18, 1959). According to A. B. Baker, Chairman of the AAN Committee on Special Courses, 6 proved popular courses are to be repeated; 3 new courses have been added, and a special course on common neurologic syndromes has been prepared for practitioners in the Los Angeles area. The course for general practitioners is not open to AAN members.... The regular 6 courses to be repeated are neuropharmacology (April 13, 1959), Carl C. Pfeiffer, chairman; neurophysiology (April 13, 1959), James L. O'Leary, chairman; neurochemistry (April 14, 1959), Maynard M. Cohen and Donald B. Tower, chairmen; neuropathology (April 14, 1959), Erland R. Nelson, chairman; neuroroentgenology (April 14, 1959), A. L. Sahs, chairman, and Harold Peterson, lecturer; and electroencephalography (April 15, 1959), Frank Morrell, chairman. The new courses offered for the first time are in neuroendocrinology, neuritis, and the neuralgias and on special diagnostic procedures. The course in neuroendocrinology (April 13, 1959), under Joe R. Brown, is designed to demonstrate the close relationship between the nervous and endocrine systems, a growing field of importance to clinical neurologists. To be reviewed are important experimental advances in neuroendocrinology and the clinical aspects of neuroendocrine dysfunction of significance to neurologists. The course in neuritis and the neuralgias, under the direction of Charles A. Kane, will review the recent advances in the diagnosis and management of these syndromes. The course on special diagnostic procedures (April 15, 1959), under Nathan S. Schlezinger, will demonstrate special procedures which facilitate the neurological work-up and the practice of neurology. Among these are electromyography, biochemical procedures, isotope technics, neuroophthalmology, and neurootology with special reference to the testing of vestibular and eighth nerve functions.... The fees for all courses except neuropathology are $10 for junior members, $15 for fellows, actives, and associates, and $20 for nonmembers. Fees for the course in neuropathology are $15 for juniors, $20 for fellows, actives, and associates, and $30 for nonmembers. In addition, this course requires a $25 deposit on slides to be refunded when the slides are returned. A set of 100 specially prepared microscopic slides with appropriate legends will be forwarded to each registrant two months before the course. Selected slides

from the set will be demonstrated and discussed on the evening of the course. Thus, each applicant will have an opportunity to study the slides both before and during the course. Plastic-embedded gross specimens of common neuropathologic lesions will be on display for study throughout the duration of the course.... Since enrollment in all courses is limited, early submission of applications is strongly urged. Additional course outlines and application forms may be obtained from Mrs. J. C. McKinley, AAN Executive Secretary, 4307 East 50th Street, Minneapolis 17. All completed application forms should be sent to Mrs. McKinley.

Special Symposium for Neuroanatomists

The second annual meeting of the AAN Section of Neuroanatomical Sciences will take place (April 16-17, 1959) at the annual Academy meeting in the Statler-Hilton in Los Angeles Friday morning (April 17) has been set aside for a special symposium on the rhinencephalon and Thursday afternoon (April 16) will be used for the presentation of general scientific papers.... The Secretary of the AAN Section of Neuroanatomical Sciences is Dr. Ray S. Snider, Northwestern University, 303 East Chicago Avenue, Chicago 11.

Neuromuscular Disorders Featured in Program of ARNMD

Though handicapped by the sudden untimely death (Nov. 19, 1958) of its president, Lee M. Eaton, the scientific sessions of the Association for Research in Nervous and Mental Diseases highlighted current research in neuromuscular disorders at its Thirty-Eighth Annual Meeting in the Hotel Roosevelt, New York City (December 12-13, 1958). The two-day concentrated sessions were divided into 5 parts. On the first day (December 12), Part I of the program considered the basic structure and function of the motor unit with formal presentations by Fritz Buchthal (general concepts), A. J. Hodge (ultrastructure), John Gergely (biochemistry of muscular contraction), and Jose Del Castillo (neuromuscular transmission). Part II was devoted to experimental pathology with contributions by D. Denny-Brown (experimental studies on atrophy, hypertrophy, degeneration, and regeneration) and by Betty Q. Banker (experimental myopathies). Part III considered basic approaches to clinical problems of neuromuscular disorders in man with presentations by E. H. Lambert (neurophysiologic technics), G. Milton Shy (endocrine and metabolic studies), and Raymond D. Adams (histopathologic studies).... On the second day (December 13), Part IV of the program was devoted to clinical problems with contributions by J. N. Walton (muscular dystrophy), C. M. Pearson and A. S. Rose (the myositides), P. R. Dodge (congenital musculoskeletal disorders), J. A. Bastron and E. H. Lambert (the myotonias), L. P. Rowland, P. F. A. Hoefer, and H. Aranow, Jr. (myasthenic syndromes), David Grob and R. J. Johns (transient paralysis), G. Wohlfart and I. Gamstorp, both of Lund, Sweden (chronic progressive lower motor neuron disorders), and D. N. Buchanan (flaccidity in childhood). Part V or the final

section of the program delineated promising experimental technics in neuromuscular disorders with contributions by H. Herrmann (tissue culture), R. J. Johns and David Grob (micro-electrode studies), T. W. Farmer and F. Buchthal (refractory and irresponsive periods), E. Ronzoni, L. Berg, and W. M. Landau (enzyme studies of biopsy material), Igor Klatzo (immunochemistry), and Beni Horvath (structure and chemistry of proteins in muscle disease).... Other members of the ARNMD Program Committee, in addition to its recently deceased Chairman, Lee M. Eaton,* were Raymond D. Adams, D. Denny-Brown, Edward H. Lambert, and G. Milton Shy.

Sidney Farber Wins Weinstein Award

On Saturday evening, November 15, Academy member, Sidney Farber, received the 1958 Max Weinstein Award for his leadership and contributions to cerebral palsy research. The ceremony was performed at the President's Dinner, following the conclusion of the Ninth Annual Conference of the United Cerebral Palsy Associations, held at the Mayflower Hotel, Washington, D. C. (November 14-16, 1958). The presentation of the Weinstein Plaque and a check of $1,000 to Dr. Farber was made by H. Houston Merritt, Chairman of the UCP Medical and Scientific Committee, who recently was elected a vice-president of the organization. After an eloquent response by Dr. Farber, Charles S. Cameron, Dean of the Hahnemann Medical College, spoke on "The Future of Medical Research in the United States" and UCP President Roger S. Firestone delivered a stirring presidential address. The festivities ended with dancing to the music of Jack Morton's Orchestra.

Neurology-Neurosurgery Unit Inaugurated in Mexico City

On November 18, 1958, the new Neurology-Neurosurgery Unit of the Mexico City General Hospital was inaugurated by the President of the Mexican Republic, Don Adolfo Ruiz Cortines, accompanied by Dr. Ignacio Morones Prieto, the incumbent Secretary of Health and Public Welfare. The official ceremonies were opened with an address by Dr. Clemente Robles, the head of the new neurology unit, who traced the history of Mexican medical advances from the time of the first field hospital established by the Conquistador Hernan Cortes through the development of the present General Hospital† which was founded by Porfirio Diaz in 1911. He concluded by stating that the opening of the new Neurology-Neurosurgery Unit, the only one of its kind in the country, represented an important forward step in the study and treatment of neurological diseases in Mexico. Following other speeches by Dr. Leonides Guadarrama, Director of the General Hospital, and by President Cortines himself, the President and his retinue toured the neurological-neurosurgical unit. The new unit has 42 beds for adult patients, two small pediatric wards, and surgical, EEG, and x-ray suites. It

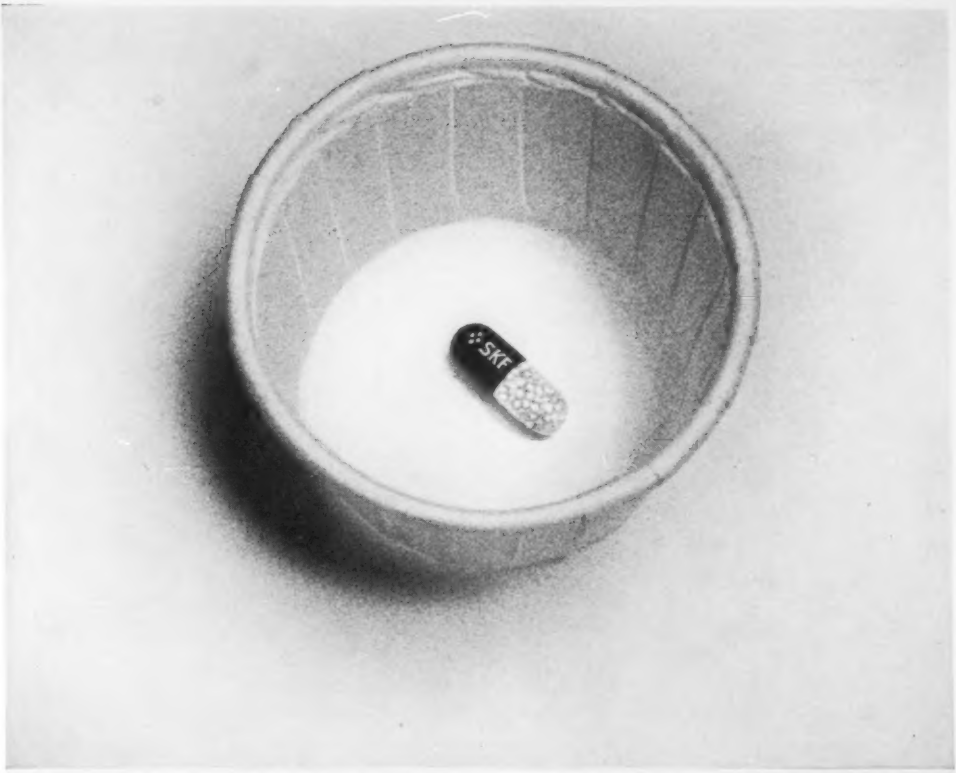
*Dr. Eaton also was the 1958 ARNMD President and Chairman of its Commission.
†Adjacent to the General Hospital are the Children's Hospital (1944), the National Institute of Cardiology (1946), the Hospital for Nutritional Diseases (1947), and the Neurology-Neurosurgery Unit. In this Medical Center is concentrated most of Mexico's medical research.

is furnished with new and modern equipment and ready for occupancy by patients. The cost of construction was 3 million pesos.... Present at the inaugural ceremonies were official guests from other countries. From the U.S.A. came Derek Denny-Brown, Pearce Bailey, John F. French, and Webb Haymaker; from France, Prof. and Madame Paul Dell and Dr. Antoine Remond; from Chile, Raul Hernandez Peon; and from Cuba, Dr. C. M. Ramirez Corria. The day (November 19) following the inaugural ceremonies, the visiting neurologists and neurosurgeons joined their Mexican colleagues in a three-day scientific program (November 19-21) held in the General Hospital and at the National University. Among the Mexican participants were Drs. Luis Saenz-Arroyo, Manuel Velasco-Svarez (President of the Mexican Society of Neurosurgery), Hernando Guzman West (President of the newly formed Mexican Society of Neurology and Psychiatry), Roberto Gamboa Acosta, Ramon del Cueto, Guillermo Santin, Mariano Vazquez, and Armando Ortiz-Galvan, currently a Research Associate of Maitland Baldwin at NINDB, who, with Jose Humberto Mateos, were of great help in guiding the visiting groups.

Richard P. Schmidt To Develop Neurology Program in Gainesville

Richard P. Schmidt has moved from the University of Washington in Seattle to the University of Florida College of Medicine in Gainesville. He has accepted the task of developing there a program in academic neurology. He writes, "Initially, I am setting up my own research laboratories and getting started with a teaching plan for the medical students. I eventually hope to have a residency training program for the training of academic neurologists as well as practitioners of neurology."

Pearce Bailey



Better protection against relapse with Compazine* Spansule† capsules

With 'Compazine' Spansule capsule therapy you maintain firmer and more continuous control of psychotic behavior than with usual tablet therapy

Hard-won control of psychotic behavior is too often jeopardized by sudden falls in drug levels due to skipped or forgotten doses of t.i.d. and q.i.d. tablet medication.

Because one 'Spansule' capsule sustains an unvarying therapeutic level of 'Compazine' for 10 to 12 hours, 'Compazine' *Spansule* capsule therapy protects your patient from these sudden falls in drug levels that may lead to a breakthrough of symptoms.

Administered only q12h, 'Compazine' *Spansule* capsules help you maintain continuous, all-day and all-night control of psychotic behavior.

Available: NEW 75 mg. 'Compazine' *Spansule* capsules—also in strengths of 10 mg., 15 mg. and 30 mg.

If it's a 'Spansule' brand capsule, it's made and marketed only by

Smith Kline & French Laboratories

first ⧗ in sustained release oral medication

*T.M. Reg. U.S. Pat. Off. for prochlorperazine, S.K.F.
†T.M. Reg. U.S. Pat. Off. for sustained release capsules, S.K.F.

UNIVERSITY OF TORONTO

FACULTY OF MEDICINE

POSTGRADUATE COURSE IN ELECTROENCEPHALOGRAPHY

A full time course extending over a period of one year is offered. The first six months will be directed to the theory and practice of electroencephalography. The second six months will be devoted to obtaining practical experience. During the first six months, seminars on Neuroanatomy, Neurophysiology and Neuropathology and the theory and practice of Electroencephalography will be offered with interpretation of records in the University Teaching Hospitals. The curriculum fulfills the requirements of the Board of Qualifications of the American Electroencephalographic Society.

The course will commence on September 1st, 1959 and will be limited to twelve candidates. Only six candidates can be accepted for the six months of practical experience. The fee for the six months' didactic portion of the course will be $500.00 (Canadian Funds), and registration as a postgraduate student, with a fee of $5.00, for the remaining six months of the course in Toronto. The second portion of the course cannot be taken before completion of the first.

For further information and applications write to

Division of Postgraduate Medical Education, Faculty of Medicine, University of Toronto, Toronto 5, Canada

HALL-BROOKE

An Active Treatment Hospital, located one hour from New York.

A private hospital devoted to active treatment, analytically-oriented psychotherapy, and the various somatic therapies.

HALL-BROOKE,
Greens Farms, Box 31, Conn. Telephone: Westport CApital 7-1251

GEORGE S. HUGHES, M.D.
LEO H. BERMAN, M.D.
ALBERT M. MOSS, M.D.
LOUIS J. MICHEELS, M.D.
ROBERT ISENMAN, M.D.
JOHN D. MARSHALL, JR., M.D.
PETER P. BARBARA, Ph.D.

complete integrated facilities for

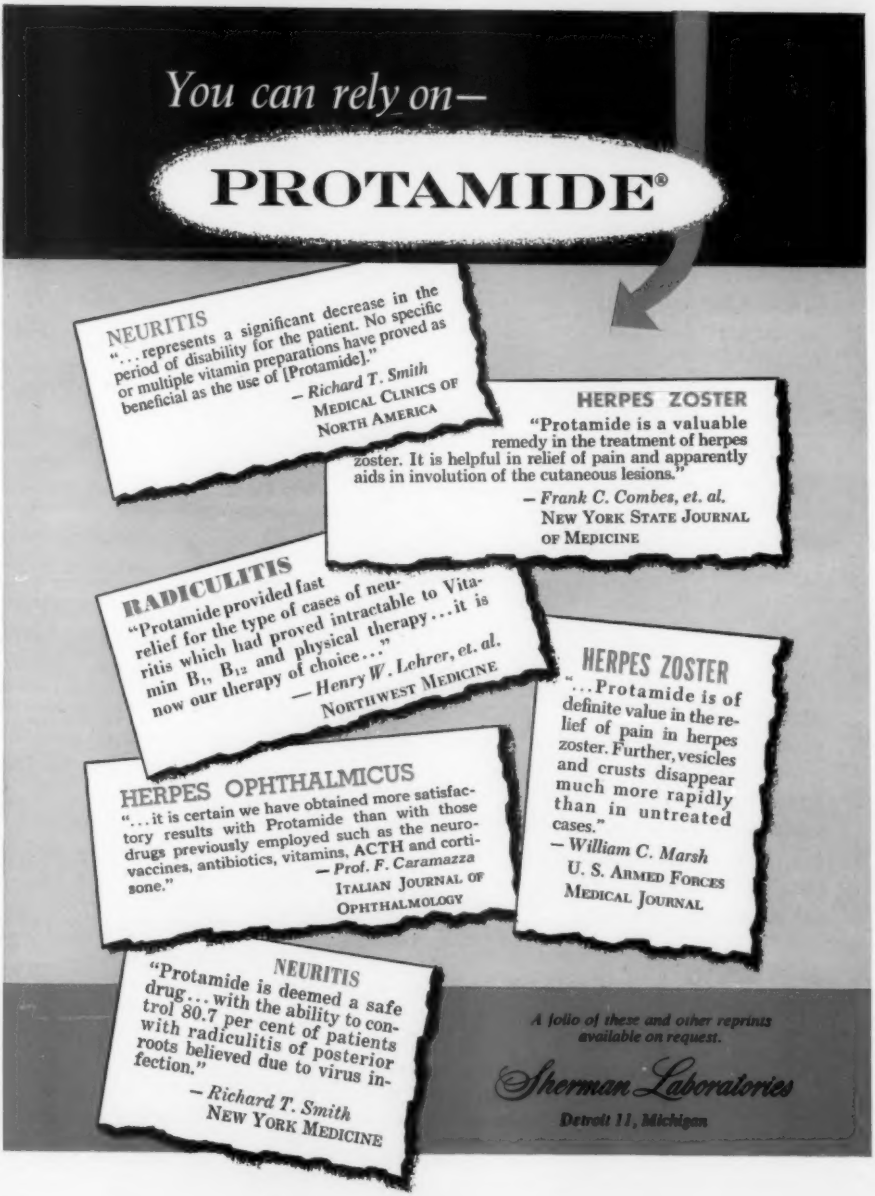
ELECTROMYOGRAPHY

- **SINGLE CHANNEL EMG** with two-channel magnetic tape recorder for recording notes and EMG simultaneously. Model TE 1.2-7
- **TWO-CHANNEL EMG** permits simultaneous recording and study of two EMG potentials or of one potential together with a related physical parameter such as force or pressure. Model TE 2-7

• Automatic controls and new circuits provide simplified reliable operation without shielded rooms in most locations. • EMG potentials faithfully reproduced by special recorder circuits. • Specifications equal or surpass requirements for research, teaching and clinical use.

NEW rugged COAXIAL NEEDLE ELECTRODE withstands autoclaving, has tapered shaft, 26 gauge at tip, insulated handle.

TECA CORPORATION
80 Main St.
White Plains
New York


You can rely on–
PROTAMIDE®
NEURITIS
"... represents a significant decrease in the period of disability for the patient. No specific or multiple vitamin preparations have proved as beneficial as the use of [Protamide]."
– Richard T. Smith
MEDICAL CLINICS OF NORTH AMERICA
HERPES ZOSTER
"Protamide is a valuable remedy in the treatment of herpes zoster. It is helpful in relief of pain and apparently aids in involution of the cutaneous lesions."
– Frank C. Combes, et. al.
NEW YORK STATE JOURNAL OF MEDICINE
RADICULITIS
"Protamide provided fast relief for the type of cases of neuritis which had proved intractable to Vitamin B1, B12 and physical therapy... it is now our therapy of choice..."
– Henry W. Lehrer, et. al.
NORTHWEST MEDICINE
HERPES ZOSTER
"...Protamide is of definite value in the relief of pain in herpes zoster. Further, vesicles and crusts disappear much more rapidly than in untreated cases."
– William C. Marsh
U. S. ARMED FORCES MEDICAL JOURNAL
HERPES OPHTHALMICUS
"...it is certain we have obtained more satisfactory results with Protamide than with those drugs previously employed such as the neurovaccines, antibiotics, vitamins, ACTH and cortisone."
– Prof. F. Caramazza
ITALIAN JOURNAL OF OPHTHALMOLOGY
NEURITIS
"Protamide is deemed a safe drug... with the ability to control 80.7 per cent of patients with radiculitis of posterior roots believed due to virus infection."
– Richard T. Smith
NEW YORK MEDICINE
A folio of these and other reprints available on request.
Sherman Laboratories
Detroit 11, Michigan

This myelogram, made with the spot-film device, shows a herniated sixth cervical intervertebral disk. A concave filling defect of the *Pantopaque* column in the subarachnoid space is present, extending from the midportion of the body of the sixth cervical vertebra to the lower margin of the seventh cervical vertebra on the left side. The sheath of the nerve root between the sixth and seventh cervical vertebrae is obliterated, and the root sheath between the seventh cervical and first thoracic vertebrae is deformed. The cervical spinal cord is displaced slightly toward the right.

"PANTOPAQUE" *is the registered trademark under which all leading x-ray dealers supply the compound ethyl iodophenylundecylate, which is synthesized by the Research Laboratories of Eastman Kodak Company and prepared as the myelographic contrast medium Iodophenylate Injection, U.S.P., by Lafayette Pharmacal Company. The trademark serves to indicate to the radiologist continuity of experience in the manufacture of this medium.*

RATED FIRST
FOR TRANQUILIZING EFFECT[1]

IN HOSPITALIZED PSYCHIATRIC PATIENTS, Miltown has demonstrated great usefulness in relieving anxiety and tension.[1-5]

In tranquilizing effect Miltown has been found superior to phenothiazine derivatives and Rauwolfia products.[1] However, its anti-psychotic effect is less pronounced than that of the other drugs.[1]

On the other hand, combined with an effective anti-psychotic preparation, Miltown becomes "extremely valuable in alleviating the overactivity, tension, excitement and anxiety of the psychotic."[1]

An added advantage of Miltown is relaxation of skeletal muscle, not obtained with most other tranquilizers.

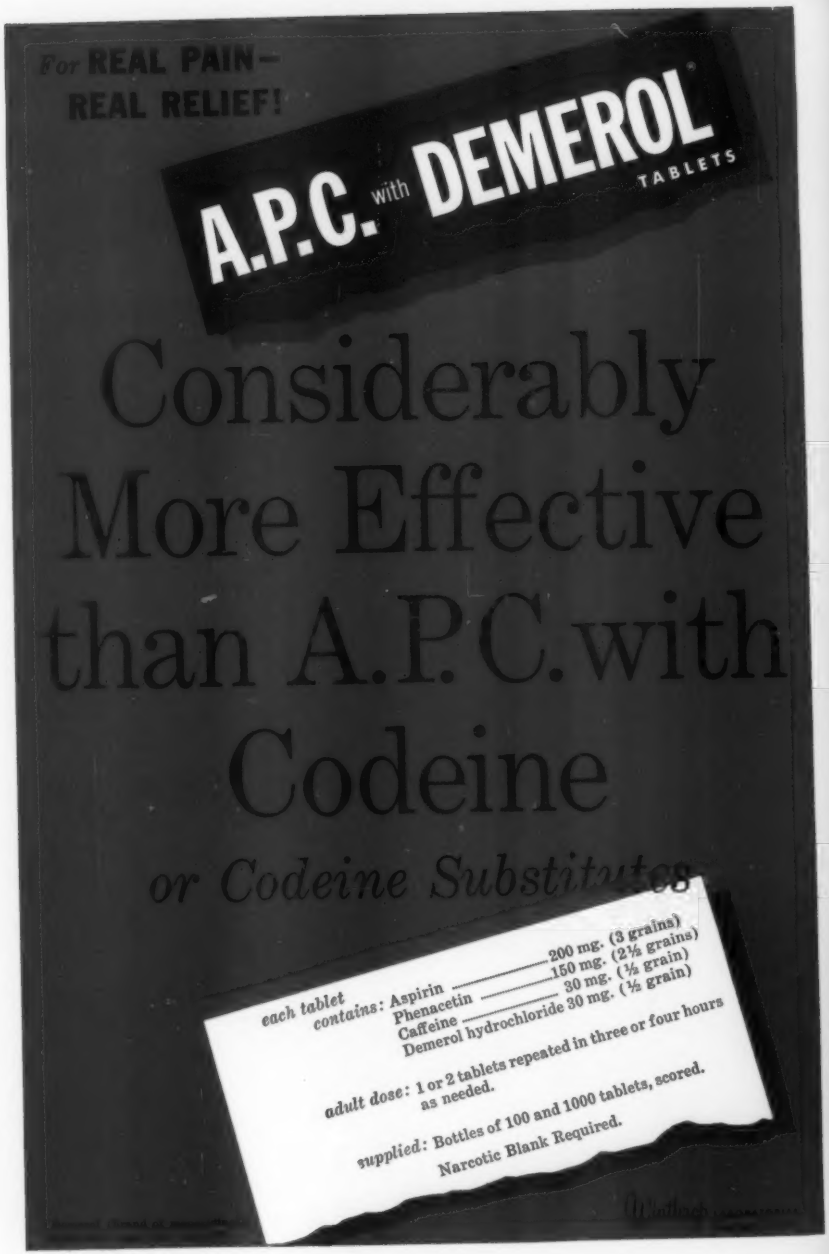
References: 1. Barsa, J. A.: Am. J. Psychiat. 115:79, July 1958. 2. Graffagnino, P. N., Friel, P. B. and Zeller, W. W.: Connecticut M. J. 21:1047, Dec. 1957. 3. Hollister, L. E., Elkins, H., Hiler, E. G. and St. Pierre, R.: Ann. New York Acad. Sc. 67:789, May 9, 1957. 4. Pennington, V. M.: Am. J. Psychiat. 114:257, Sept. 1957. 5. Tucker, K. and Wilensky, H.: Am. J. Psychiat. 113:698, Feb. 1957.

Available in 400 mg. scored and 200 mg. sugar-coated tablets. Also available as MEPROSPAN* (200 mg. meprobamate *continuous release* capsules). *TRADE-MARK

Miltown®
meprobamate (Wallace)

CM-7801

WALLACE LABORATORIES,
New Brunswick, N. J.


For REAL PAIN—
REAL RELIEF!
A.P.C. with DEMEROL®
TABLETS
Considerably
More Effective
than A.P.C. with
Codeine
or Codeine Substitutes
each tablet
contains: Aspirin ..... 200 mg. (3 grains)
Phenacetin ..... 150 mg. (2½ grains)
Caffeine ..... 30 mg. (½ grain)
Demerol hydrochloride 30 mg. (½ grain)
adult dose: 1 or 2 tablets repeated in three or four hours as needed.
supplied: Bottles of 100 and 1000 tablets, scored.
Narcotic Blank Required.